adenosine-mediated immunosuppressive effects play a role. Hence, there is a need for treatments of such diseases and conditions.

SUMMARY

Provided herein are variant Adenosine Deaminase 2 (ADA2) proteins or catalytically active portions thereof that contain a modification(s) in the sequence of amino acids of an unmodified ADA2 protein or a catalytically active portion thereof. In some embodiments, the unmodified ADA2 protein can include the sequence of amino acids set forth in SEQ ID NO:5, or a sequence of amino acids that can exhibit at least 85% sequence identity to the sequence of amino acids set forth in SEQ ID NO:5, or is a catalytically active portion thereof; the amino acid modification(s) are selected from among amino acid replacement(s), deletion(s) and insertion(s); and the variant ADA2, when in dimer form, can exhibit one or more properties selected from among increased adenosine deaminase activity, reduced heparin binding, longer serum half-life, altered pH optimum, increased thermal stability, altered receptor binding and hyperglycosylation compared to the corresponding dimer form of the unmodified ADA2 protein. A variety of amino acid modifications, including replacements, deletions and insertions are provided. It is understood that the discreet modifications that confer a particular activity or property can be combined; as in proteins effects of mutation or modifications generally are additive. Any of the variant ADA2 or catalytically active portion thereof provided herein that contains modifications, including replacements, deletions and insertions, and nucleic acids encoding the variant ADA2 or catalytically active portion thereof, can be used in any of the methods, compositions, conjugates, modified forms, vectors, cells, combinations, uses and compositions for use, and combinations for use, provided herein.

In some embodiments, there is provided 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:

the unmodified ADA2 protein comprises the sequence of amino acids set forth in SEQ ID NO:5, or a sequence of amino acids that exhibits at least 95% sequence identity to the sequence of amino acids set forth in SEQ ID NO:5, or is a catalytically active portion thereof;

the amino acid modification(s) are selected from among amino acid replacement(s), deletion(s) and insertion(s);

the variant ADA2 protein, when in dimer form, exhibits one or more properties selected from among increased adenosine deaminase activity, reduced heparin binding, longer serum half-life, altered pH optimum, increased thermal stability, altered receptor binding and hyperglycosylation 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; and

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

In some embodiments, there is provided a variant Adenosine Deaminase 2 (ADA2) protein or a catalytically active portion thereof as described herein, comprising a modification(s) in the sequence of amino acids of an unmodified ADA2 polypeptide or a catalytically active portion thereof, wherein:

the unmodified ADA2 protein is selected from among an ADA protein that comprises:

the sequence of amino acids set forth in SEQ ID NO:5;, or

a sequence of amino acids that exhibits at least 95% sequence identity to the sequence of amino acids set forth in SEQ ID NO:5, or is a catalytically active portion of the sequence of amino acids that exhibits at least 95% sequence identity to the sequence of amino acids set forth in SEQ ID NO:5; or

comprises the sequence of amino acids set forth in any of SEQ ID NOS: 5, 326-334, 340, 375 and 380-383 or is a catalytically active portion thereof;

the modified ADA protein has up to 2, 3, 4, 5, 6, 7, 8, 9 or 10 amino acid modifications compared to the unmodified ADA protein;

the amino acid modification(s) are selected from among amino acid replacement(s), deletion(s) and insertion(s);

the variant ADA2 protein comprises an amino acid replacement at one or both of positions corresponding to amino acid residue 219 and 262 with reference to amino acid positions set forth in SEQ ID NO:5.

In some embodiments, there is provided a catalytically active portion of the variant ADA2 protein as described herein, wherein the unmodified ADA2 protein is a homodimer of corresponding catalytically active portions of the polypeptide whose sequence is set forth in of SEQ ID NO:5, wherein corresponding portions are determined by alignment.

In some embodiments, there is provided a catalytically active portion of the variant ADA2 protein or catalytically active portion thereof as described herein, wherein the catalytically active portion comprises the ADA domain.

In some embodiments, there is provided a variant ADA2 multimer, comprising a plurality of variant ADA2 proteins or catalytically active portions thereof as described herein, wherein the variant ADA2 protein or catalytically active portion thereof are the same or different.

In some embodiments, there is provided a variant ADA2 dimer, comprising a variant ADA2 protein or catalytically active portion thereof as described herein.

In some embodiments, there is provided a conjugate, comprising an ADA2 protein or catalytically active portion thereof as described herein, linked directly or indirectly via a linker to a heterologous moiety, wherein the heterologous moiety is a half-life extending moiety.

In some embodiments, there is provided a nucleic acid encoding a variant ADA2 protein or catalytically active portion thereof as described herein.

In some embodiments, there is provided a vector, comprising the nucleic acid molecule as described herein.

In some embodiments, there is provided an isolated cell or a cell culture, comprising the vector as described herein.

In some embodiments, there is provided a method of producing an ADA2 variant or catalytically active portion thereof, comprising culturing a cell as described herein, under conditions whereby the ADA2 variant is expressed.

In some embodiments, there is provided a method, comprising, culturing or expanding the cells as described herein to produce expanded cells.

In some embodiments, there is provided use of the expanded cells as described herein for manufacture of a medicament treatment of a tumor or cancer.

In some embodiments, there is provided a pharmaceutical composition, comprising a variant ADA2 protein as described herein or a catalytically active portion thereof, or a dimer or multimer of the ADA2 protein or catalytically active portion thereof or a conjugate as described herein.

In some embodiments, there is provided an ADA variant or multimer thereof as described herein or the conjugate as described herein for treatment of a tumor, cancer, non-cancer hyperproliferative disease, a fibrotic disease, an infectious disease, a vasculopathy, or an immunodeficiency disease.

In some embodiments, there is provided use of pharmaceutical composition as described herein for manufacture of a medicament treatment of a tumor, cancer, non-cancer hyperproliferative disease, a fibrotic disease, an infectious disease, a vasculopathy, or an immunodeficiency disease in a subject.

In some embodiments, the variant ADA2 protein or catalytically active portion thereof, when in dimer form, exhibits increased adenosine deaminase activity or increased adenosine deaminase activity and reduced heparin binding.

In some embodiments, the unmodified ADA2 protein is a homodimer, and the monomer form comprises the sequence of amino acid residues set forth in SEQ ID NO:5. In some embodiments, the variant ADA2 is a catalytically active portion of the variant ADA2 protein as provided herein, wherein the unmodified ADA2 protein is a homodimer of corresponding catalytically active portions of the polypeptide whose

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:

the unmodified ADA2 protein comprises the sequence of amino acids set forth in SEQ ID NO:5, or a sequence of amino acids that exhibits at least 95% sequence identity to the sequence of amino acids set forth in SEQ ID NO:5, or is a catalytically active portion thereof;

the amino acid modification(s) are selected from among amino acid replacement(s), deletion(s) and insertion(s);

the variant ADA2 protein, when in dimer form, exhibits one or more properties selected from among increased adenosine deaminase activity, reduced heparin binding, longer serum half-life, altered pH optimum, increased thermal stability, altered receptor binding and hyperglycosylation 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; and

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

2. The variant ADA2 protein of claim 1, wherein the variant ADA2 protein comprises one or more amino acid replacement(s) at an amino acid position corresponding to amino acid residue 11, 20, 109, 118, 119, 124, 133, 139, 183, 191, 219, 221, 224, 262, 264, 366, 371, 372 or 452, with reference to amino acid positions set forth in SEQ ID NO:5.

3. The variant ADA2 protein or catalytically active portion thereof claim 1, wherein:

the modification(s) is on or more amino acid replacement(s); and

the variant ADA2 protein comprises one or more amino acid replacement(s) at an amino acid position corresponding to amino acid residue 11, 13, 20, 22, 26, 86, 179, 217, 219, 221, 258, 262, 264, 266, 267, 277, 283, 296, 309, 317, 321, 352, 366, 371, 372, 373, 374, 403, 404, 405, 406, 441, 444, 452, 461, 469 or 470, with reference to amino acid positions set forth in SEQ ID NO:5.

4. The variant ADA2 protein or catalytically active portion thereof of any of claims 1-3, wherein the ADA2 protein does not contain a modification selected from among an amino acid replacement corresponding to H7R, G18A, G18R, G18V, I64T, A80D, H83Q, V90A, C108G, A120V, H121R, W133G, R125C, R140Q, K141R, R142W, P164L, P222L, W235S, H306R, E330G, W333G,

V365L, Y424C, F464S or a deletion corresponding to R8-K14del→--, with numbering with reference to amino acid residues set forth in SEQ ID NO:5.

5. The variant ADA2 protein or catalyically active portion thereof of any of claims 1-4, wherein the unmodified ADA2 protein is selected from among an ADA protein that comprises:

the sequence of amino acids set forth in SEQ ID NO:5; or

a sequence of amino acids that exhibits at least 95% sequence identity to the sequence of amino acids set forth in SEQ ID NO:5, or is a catalytically active portion of the sequence of amino acids that exhibits at least 95% sequence identity to the sequence of amino acids set forth in SEQ ID NO:5; or

the sequence of amino acids set forth in any of SEQ ID NOS: 5, 326-334, 340, 375 and 380-383 or is a catalytically active portion thereof;.

6. The variant ADA2 protein or catalytically active portion thereof of any of claims 1-5 or catalytically active portion thereof, wherein the variant ADA2 protein, when in dimer form, exhibits reduced heparin binding compared to the ADA2 protein of SEQ ID NO:5 or the corresponding catalytically active portion thereof.

7. The variant ADA2 protein of any of claims 1-6 , when in dimer form, exhibits increased adenosine deaminase activity or 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; and

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

8. The variant ADA2 protein or catalytically active portion thereof of any of claims 1-6, wherein the variant ADA2 protein comprises one or more amino acid replacement(s) selected from among R20A, R20D, R20E, S262N, R366A, R366D, R366E, K371A, K371D, K371E, K372A, K372D, K372E and K452E, with reference to amino acid positions set forth in SEQ ID NO:5.

9. The variant ADA2 protein or catalytically active portion thereof of any of claims 1- 7, comprising one or more amino acid replacement(s) corresponding to K11A; K11D; K11E; K13A; K13D; K13E; K371A; K371D; K371E; K372A; K372D; K372E; K452A; K452D; K452E; R20A; R20D; R20E; R366A; R366D; R366E; K26A; K26D; K26E; R217A; R217D; R217E; K258A; K258D; K258E; R277A; R277D; R277E; R283A; R283D; R283E; K309A; K309D; K309E; K317A; K317D; K317E; K321A; K321D; K321E; R352A; R352D; R352E; R441A; R441D; R441E; K444A; K444D; K444E; K461A; K461D; K461E; K469A; K469D; K469E; K470A; K470D; and K470E.

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

the unmodified ADA2 protein is selected from among an ADA protein that comprises:

the sequence of amino acids set forth in SEQ ID NO:5; or

a sequence of amino acids that exhibits at least 95% sequence identity to the sequence of amino acids set forth in SEQ ID NO:5, or is a catalytically active portion of the sequence of amino acids that exhibits at least 95% sequence identity to the sequence of amino acids set forth in SEQ ID NO:5; or

comprises the sequence of amino acids set forth in any of SEQ ID NOS: 5, 326-334, 340, 375 and 380-383 or is a catalytically active portion thereof;

the modified ADA protein has up to 2, 3, 4, 5, 6, 7, 8, 9 or 10 amino acid modifications compared to the unmodified ADA protein;

the amino acid modification(s) are selected from among amino acid replacement(s), deletion(s) and insertion(s);

the variant ADA2 protein comprises an amino acid replacement at one or both of positions corresponding to amino acid residue 219 and 262 with reference to amino acid positions set forth in SEQ ID NO:5。

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

12. A catalytically active portion of the variant ADA2 protein of claim 10 or claim 11, wherein the unmodified ADA2 protein is a homodimer of corresponding catalytically active portions of the polypeptide whose sequence is set forth in of SEQ ID NO:5, wherein corresponding portions are determined by alignment.

13. variant ADA2 protein of any of claims 10-12, wherein the unmodified ADA2 protein comprises the sequence of amino acids set forth in any of SEQ ID NOS: 5, 326-330 and 380-383or is a catalytically active portion thereof.

14. The variant ADA2 protein of any of claims 10-12, wherein the unmodified ADA2 protein consists of the sequence of amino acids set forth in any of SEQ ID NOS: 5, 326-330 and 380-383 or is a catalytically active portion thereof.

15. The variant ADA2 protein of any of claims 10-13, wherein the unmodified ADA2 protein comprises the sequence of amino acids set forth in SEQ ID NO: 5 or a catalytically active portion thereof.

16. The variant ADA2 protein or catalytically active portion thereof of any of claims 10-15, wherein the variant ADA2 protein comprises one or more amino acid replacement(s) selected from among replacements corresponding to R219K; R219Q; R219N; R219A; S262A; S262V; S262M; S262N; S262C; S262D; S262E; S262F; S262G; S262H; S262I; S262K; S262L; S262P; S262Q; S262R; S262T; S262W and S262Y.

17. The variant ADA2 protein or catalytically active portion thereof of any of claims 10-15, comprising the replacement corresponding to S262N or S262Q.

18. The variant ADA2 protein or catalytically active portion thereof of any of claims 10-15, comprising the replacement corresponding to R219K, R219Q, R219N or R219A.

19. The variant ADA2 protein or catalytically active portion thereof of any of claims 10-15, comprising the replacement corresponding to R219Q/S262N.

20. The variant ADA2 protein or catalytically active portion thereof of claim 19, selected from among any of R219Q/S262N/R20N/V22S, R219Q/S262N/K371N/D373S, R219Q/S262N/K372N/I374S, R219Q/S262N/T403N/H405S, R219Q/S262N/G404N/P406S, R219Q/S262N/C105-T147del→(Gly)15, R219Q/S262N/C105-T147del→(Gly)10, R219Q/S262N/C105-T147del→(Gly)7, R219Q/S262N/C105-T147del→(Gly)5, R219Q/S262N/C105-T147del→(Gly)3, R219Q/S262N/R125N/P126A, R219Q/S262N/S127N/K129S, R219Q/S262N/P126N/E128T, R219Q/S262N/R112N/I114T, R219Q/S262N/I134N/L135C/L136T, R219Q/S262N/I134N/L135S/L136T, R219Q/S262N/R142N/Q144S, R219Q/S262N/E137N/Y139T, R219Q/S262N/P111N/G113S, R219Q/S262N/F119S, R219Q/S262N/F119K, R219Q/S262N/Y224R, R219Q/S262N/Y224N, R219Q/S262N/Y191S, R219Q/S262N/Y191D, R219Q/S262N/F183K, R219Q/S262N/Y191D/Y224R, R219Q/S262N/F109S, R219Q/S262N/F109A, R219Q/S262N/R118D, R219Q/S262N/R118A, R219Q/S262N/Y139T, R219Q/S262N/Y139A, R219Q/S262N/W133S, R219Q/S262N/W133T, R219Q/S262N/P124A, R219Q/S262N/P124S, R219Q/S262N/V99-Q144del→$(GGGGS)_1$, R219Q/S262N/V99-Q144del→$(GGGGS)_2$, R219Q/S262N/V99-Q144del→$(GGGGS)_3$, R219Q/S262N/C105-T147del→$(GGGGS)_1$, R219Q/S262N/C105-T147del→$(GGGGS)_2$, R219Q/S262N/C105-T147del→$(GGGGS)_3$, R219Q/S262N/K371D/V99-Q144del→$(GGGGS)_1$, R219Q/S262N/K371D/V99-Q144del→$(GGGGS)_2$, R219Q/S262N/K371D/V99-Q144del→$(GGGGS)_3$, R219Q/S262N/K371D/C105-T147del→$(GGGGS)_1$,

R219Q/S262N/K371D/C105-T147del→$(GGGGS)_2$, R219Q/S262N/K371D/C105-T147del→$(GGGGS)_3$, R219Q/S262N/C105-T147del→$(Gly)_n$, R219Q/S262N/K11A, R219Q/S262N/K11D, R219Q/S262N/K11E, R219Q/S262N/K13A, R219Q/S262N/K13D, R219Q/S262N/V99-Q144del→$(GGGGS)_n$, R219Q/S262N/C105-T147del→$(GGGGS)_n$, R219Q/S262N/N98-N156del, R219Q/S262N/C105-E148del, R219Q/S262N/C105-T147del, R219Q/S262N/V99-Q144del, R219Q/S262N/K371D/C105-T147del→$(Gly)_n$, R219Q/S262N/K371D/C105-T147del→$(Gly)_{15}$, R219Q/S262N/K371D/C105-T147del→$(Gly)_{10}$, R219Q/S262N/K371D/C105-T147del→$(Gly)_7$, R219Q/S262N/K371D/C105-T147del→$(Gly)_5$, R219Q/S262N/K371D/C105-T147del→$(Gly)_3$, R219Q/S262N/K371D/V99-Q144del→$(GGGGS)_n$, R219Q/S262N/K371D/C105-T147del→$(GGGGS)_n$, R219Q/S262N/K371D/N98-N156del, R219Q/S262N/K371D/C105-E148del, R219Q/S262N/K371D/C105-T147del, R219Q/S262N/K371D/V99-Q144del, R219Q/S262N/K13E, R219Q/S262N/K371A, R219Q/S262N/K372A, R219Q/S262N/K372D, R219Q/S262N/K372E, R219Q/S262N/K452A, R219Q/S262N/K452D, R219Q/S262N/K452E, R219Q/S262N/R20A, R219Q/S262N/R20D, R219Q/S262N/R366A, R219Q/S262N/R366D, R219Q/S262N/R366E, R219Q/S262N/H264A, R219Q/S262N/H264Q, R219Q/S262N/H264N, R219Q/S262N/H264G, R219K/S262N, R219N/S262N, R219A/S262N, R219Q/S262N/L221A, R219Q/S262N/L221V, R219Q/S262N/L221G, R219Q/S262N/E179D, R219Q/S262N/E179A, R219Q/S262N/E179S, R219Q/S262N/E179T, R219Q/S262N/E179V, R219Q/S262N/E179G, R219Q/S262A, R219Q/S262V, R219Q/S262M, R219Q/S262N/K11A/R20A, R219Q/S262N/K11A/R20A/K371A, R219Q/S262N/R20A/K371A, R219Q/S262N/K11A/K371A, R219Q/S262N/K26A, R219Q/S262N/K26D, R219Q/S262N/K26E, R219Q/S262N/R217A, R219Q/S262N/R217D, R219Q/S262N/R217E, R219Q/S262N/K258A, R219Q/S262N/K258D, R219Q/S262N/K258E, R219Q/S262N/R277A, R219Q/S262N/R277D, R219Q/S262N/R277E, R219Q/S262N/R283A, R219Q/S262N/R283D, R219Q/S262N/R283E, R219Q/S262N/K309A, R219Q/S262N/K309D, R219Q/S262N/K309E, R219Q/S262N/K317A, R219Q/S262N/K317D, R219Q/S262N/K317E, R219Q/S262N/K321A, R219Q/S262N/K321D, R219Q/S262N/K321E, R219Q/S262N/R352A, R219Q/S262N/R352D, R219Q/S262N/R352E, R219Q/S262N/R441A, R219Q/S262N/R441D, R219Q/S262N/R441E, R219Q/S262N/K444A, R219Q/S262N/K444D, R219Q/S262N/K444E, R219Q/S262N/K461A, R219Q/S262N/K461D, R219Q/S262N/K461E, R219Q/S262N/K469A, R219Q/S262N/K469D, R219Q/S262N/K469E, R219Q/S262N/K470A, R219Q/S262N/K470D, R219Q/S262N/K470E, R219Q/S262N/D86A, R219Q/S262N/D86C, R219Q/S262N/D86E, R219Q/S262N/D86F, R219Q/S262N/D86G, R219Q/S262N/D86H, R219Q/S262N/D86I, R219Q/S262N/D86K, R219Q/S262N/D86L, R219Q/S262N/D86M, R219Q/S262N/D86N, R219Q/S262N/D86P, R219Q/S262N/D86Q,

R219Q/S262N/D86R, R219Q/S262N/D86S, R219Q/S262N/D86T, R219Q/S262N/D86V, R219Q/S262N/D86W, R219Q/S262N/D86Y, R219Q/S262N/E179C, R219Q/S262N/E179F, R219Q/S262N/E179H, R219Q/S262N/E179I, R219Q/S262N/E179K, R219Q/S262N/E179L, R219Q/S262N/E179M, R219Q/S262N/E179N, R219Q/S262N/E179P, R219Q/S262N/E179Q, R219Q/S262N/E179R, R219Q/S262N/E179W, R219Q/S262N/E179Y, R219C/S262N, R219D/S262N, R219E/S262N, R219F/S262N, R219G/S262N, R219H/S262N, R219I/S262N, R219L/S262N, R219M/S262N, R219P/S262N, R219S/S262N, R219T/S262N, R219V/S262N, R219W/S262N, R219Y/S262N, R219Q/S262N/L221C, R219Q/S262N/L221D, R219Q/S262N/L221E, R219Q/S262N/L221F,R219Q/S262N/L221H,R219Q/S262N/L221I, R219Q/S262N/L221K, R219Q/S262N/L221M, R219Q/S262N/L221N, R219Q/S262N/L221P, R219Q/S262N/L221Q, R219Q/S262N/L221R, R219Q/S262N/L221S, R219Q/S262N/L221T, R219Q/S262N/L221W, R219Q/S262N/L221Y, R219Q/S262C, R219Q/S262D, R219Q/S262E, R219Q/S262F, R219Q/S262G, R219Q/S262H, R219Q/S262I, R219Q/S262K, R219Q/S262L, R219Q/S262P, R219Q/S262Q, R219Q/S262R, R219Q/S262T, R219Q/S262W, R219Q/S262Y, R219Q/S262N/H264C, R219Q/S262N/H264D, R219Q/S262N/H264E, R219Q/S262N/H264F, R219Q/S262N/H264I, R219Q/S262N/H264K, R219Q/S262N/H264L, R219Q/S262N/H264M, R219Q/S262N/H264P, R219Q/S262N/H264R, R219Q/S262N/H264S, R219Q/S262N/H264T, R219Q/S262N/H264V, R219Q/S262N/H264W, R219Q/S262N/H264Y, R219Q/S262N/S266A, R219Q/S262N/S266C, R219Q/S262N/S266D, R219Q/S262N/S266E, R219Q/S262N/S266F, R219Q/S262N/S266G, R219Q/S262N/S266H, R219Q/S262N/S266I, R219Q/S262N/S266K, R219Q/S262N/S266L, R219Q/S262N/S266M, R219Q/S262N/S266N, R219Q/S262N/S266P, R219Q/S262N/S266Q, R219Q/S262N/S266R, R219Q/S262N/S266T, R219Q/S262N/S266V, R219Q/S262N/S266W, R219Q/S262N/S266Y, R219Q/S262N/K267A, R219Q/S262N/K267C, R219Q/S262N/K267D, R219Q/S262N/K267E, R219Q/S262N/K267F, R219Q/S262N/K267G, R219Q/S262N/K267H, R219Q/S262N/K267I, R219Q/S262N/K267L, R219Q/S262N/K267M, R219Q/S262N/K267N, R219Q/S262N/K267P, R219Q/S262N/K267Q, R219Q/S262N/K267R, R219Q/S262N/K267S, R219Q/S262N/K267T, R219Q/S262N/K267V, R219Q/S262N/K267W, R219Q/S262N/K267Y, R219Q/S262N/V296A, R219Q/S262N/V296C, R219Q/S262N/V296D, R219Q/S262N/V296E, R219Q/S262N/V296F, R219Q/S262N/V296G, R219Q/S262N/V296H, R219Q/S262N/V296I, R219Q/S262N/V296K, R219Q/S262N/V296L, R219Q/S262N/V296M, R219Q/S262N/V296N, R219Q/S262N/V296P, R219Q/S262N/V296Q, R219Q/S262N/V296R, R219Q/S262N/V296S, R219Q/S262N/V296T, R219Q/S262N/V296W and R219Q/S262N/V296Y.

21. The variant ADA2 protein of any of claims 10-20, wherein the variant ADA2 protein comprises amino acid replacements selected from among R219Q/S262N/K371E, R219Q/S262N/K371D, R219Q/S262N/R20E and R219Q/S262N/K371D/R20E.

22 The variant ADA2 protein or catalytically active portion thereof of any of claims 10-20 that comprises the modifications corresponding to R219Q/S262N/--→N1/--→A2/--→S3; R219Q/S262N/R20N/V22S; R219Q/S262N/K371N/D373S; R219Q/S262N/K372N/I374S; R219Q/S262N/T403N/H405S; or R219Q/S262N/G404N/P406S.

23. The variant ADA2 protein or catalytically active portion thereof of any of claims 10-20 that comprises amino acid replacements corresponding to R219Q/S262N/R125N/P126A; R219Q/S262N/S127N/K129S; R219Q/S262N/P126N/E128T; R219Q/S262N/R112N/I114T; R219Q/S262N/I134N/L135C/L136T; R219Q/S262N/I134N/L135S/L136T; R219Q/S262N/R142N/Q144S; R219Q/S262N/E137N/Y139T; or R219Q/S262N/P111N/G113S.

24 The variant ADA2 protein or catalytically active portion thereof of any of claims 10-23, further comprising a modification in the putative receptor binding domain (PRB) corresponding to one or more of the modifications selected from among: R125N/P126A; S127N/K129S; P126N/E128T; R112N/I114T; I134N/L135C/L136T; I134N/L135S/L136T; R142N/Q144S; E137N/Y139T; and P111N/G113S.

25. The variant ADA2 protein or catalytically active portion thereof of any of claims 10-23, comprising amino acid replacements selected from among replacements corresponding to R219Q/S262N/F119S; R219Q/S262N/F119K; R219Q/S262N/Y224R; R219Q/S262N/Y224N; R219Q/S262N/Y191S; R219Q/S262N/Y191D; R219Q/S262N/F183K; R219Q/S262N/Y191D/Y224R; R219Q/S262N/F109S; R219Q/S262N/F109A; R219Q/S262N/R118D; R219Q/S262N/R118A; R219Q/S262N/Y139T; R219Q/S262N/Y139A; R219Q/S262N/W133S; R219Q/S262N/W133T; R219Q/S262N/P124A; and R219Q/S262N/P124S.

26. The variant ADA2 protein or catalytically active portion thereof of of any of claims 10-23 that comprises amino acid replacements selected from among replacements corresponding to R219Q/S262N/C105-T147del→$(Gly)_{15}$; R219Q/S262N/C105-T147del→$(Gly)_{10}$; R219Q/S262N/C105-T147del→$(Gly)_7$; R219Q/S262N/C105-T147del→$(Gly)_5$; R219Q/S262N/C105-T147del→$(Gly)_3$; R219Q/S262N/V99-Q144del→$(GGGGS)_1$; R219Q/S262N/V99-Q144del→$(GGGGS)_2$; R219Q/S262N/V99-Q144del→$(GGGGS)_3$; R219Q/S262N/C105-T147del→$(GGGGS)_1$; R219Q/S262N/C105-T147del→$(GGGGS)_2$; R219Q/S262N/C105-T147del→$(GGGGS)_3$; R219Q/S262N/K371D/V99-Q144del→$(GGGGS)_1$; R219Q/S262N/K371D/V99-Q144del→$(GGGGS)_2$;

R219Q/S262N/K371D/V99-Q144del→$(GGGGS)_3$; R219Q/S262N/K371D/C105-T147del→$(GGGGS)_1$; R219Q/S262N/K371D/C105-T147del→$(GGGGS)_2$; R219Q/S262N/K371D/C105-T147del→$(GGGGS)_3$; R219Q/S262N/C105-T147del→(Gly)n, where n = 2 to 20; R219Q/S262N/V99-Q144del→(GGGGS)n, where n = 1 to 5; R219Q/S262N/C105-T147del→(GGGGS)n, where n = 1 to 5; R219Q/S262N/N98-N156del; R219Q/S262N/C105-E148del; R219Q/S262N/C105-T147del; R219Q/S262N/V99-Q144del; R219Q/S262N/K371D/C105-T147del→(Gly)n, where n = 2 to 20; R219Q/S262N/K371D/C105-T147del→$(Gly)_{15}$; R219Q/S262N/K371D/C105-T147del→$(Gly)_{10}$; R219Q/S262N /K371D/C105-T147del→$(Gly)_7$; R219Q/S262N/K371D/C105-T147del→$(Gly)_5$; R219Q/S262N/K371D/C105-T147del→$(Gly)_3$; R219Q/S262N/K371D/V99-Q144del→$(GGGGS)_n$, where n = 1 to 5; R219Q/S262N/K371D/C105-T147del→$(GGGGS)_n$, where n = 1 to 5; R219Q/S262N/K371D/N98-N156del; R219Q/S262N/K371D/C105-E148del; R219Q/S262N/K371D/C105-T147del; and R219Q/S262N/K371D/V99-Q144del, with reference to amino acid positions set forth in SEQ ID NO:5.

27. The variant ADA2 protein or catalytically active portion thereof of claim 10, wherein the variant ADA2 protein comprises the sequence of amino acids set forth in any of SEQ ID NOS:38-41, 51-54, 59-63, 145-159, 176-190 and 263-273 or a catalytically active portion thereof.

28. The variant ADA2 protein or catalytically active portion thereof of claim 10, wherein the variant ADA2 protein comprises the sequence of amino acids set forth in any of SEQ ID NOS: 596-645 and 658-931 or a catalytically active portion thereof

29. The variant ADA2 protein or catalytically active portion thereof of any of claims 10-23, comprising amino acid replacements selected from among replacements corresponding to R219Q/S262N/K371D; R219Q/S262N/R20E; R219Q/S262N/K371E/R20E; R219Q/S262N/K371D/R20E; R219Q/S262N; R219Q/S262N/K11A; R219Q/S262N/K11D; R219Q/S262N/K11E; R219Q/S262N/K13A; R219Q/S262N/K13D; R219Q/S262N/K13E; R219Q/S262N/K371A; R219Q/S262N/K372A; R219Q/S262N/K372D; R219Q/S262N/K372E; R219Q/S262N/K452A; R219Q/S262N/K452D; R219Q/S262N/K452E; R219Q/S262N/R20A; R219Q/S262N/R20D; R219Q/S262N/R366A; R219Q/S262N/R366D; R219Q/S262N/R366E; R219Q/S262N/H264A; R219Q/S262N/H264Q; R219Q/S262N/H264N; R219Q/S262N/H264G; R219K/S262N; R219N/S262N; R219A/S262N; R219Q/S262N/L221A; R219Q/S262N/L221V; R219Q/S262N/L221G; R219Q/S262N/E179D; R219Q/S262N/E179A; R219Q/S262N/E179S; R219Q/S262N/E179T; R219Q/S262N/E179V; R219Q/S262N/E179G; R219Q/S262A; R219Q/S262V; R219Q/S262M; R219Q/S262N/K11A/R20A; R219Q/S262N/K11A/R20A/K371A; R219Q/S262N/R20A/K371A; R219Q/S262N/K11A/K371A; R219Q/S262N/K26A;

R219Q/S262N/K26D; R219Q/S262N/K26E; R219Q/S262N/R217A; R219Q/S262N/R217D; R219Q/S262N/R217E; R219Q/S262N/K258A; R219Q/S262N/K258D; R219Q/S262N/K258E; R219Q/S262N/R277A; R219Q/S262N/R277D; R219Q/S262N/R277E; R219Q/S262N/R283A; R219Q/S262N/R283D; R219Q/S262N/R283E; R219Q/S262N/K309A; R219Q/S262N/K309D; R219Q/S262N/K309E; R219Q/S262N/K317A; R219Q/S262N/K317D; R219Q/S262N/K317E; R219Q/S262N/K321A; R219Q/S262N/K321D; R219Q/S262N/K321E; R219Q/S262N/R352A; R219Q/S262N/R352D; R219Q/S262N/R352E; R219Q/S262N/R441A; R219Q/S262N/R441D; R219Q/S262N/R441E; R219Q/S262N/K444A; R219Q/S262N/K444D; R219Q/S262N/K444E; R219Q/S262N/K461A; R219Q/S262N/K461D; R219Q/S262N/K461E; R219Q/S262N/K469A; R219Q/S262N/K469D; R219Q/S262N/K469E; R219Q/S262N/K470A; R219Q/S262N/K470D; R219Q/S262N/K470E; R219Q/S262N/D86A; R219Q/S262N/D86C; R219Q/S262N/D86E; R219Q/S262N/D86F; R219Q/S262N/D86G; R219Q/S262N/D86H; R219Q/S262N/D86I; R219Q/S262N/D86K; R219Q/S262N/D86L; R219Q/S262N/D86M; R219Q/S262N/D86N; R219Q/S262N/D86P; R219Q/S262N/D86Q; R219Q/S262N/D86R; R219Q/S262N/D86S; R219Q/S262N/D86T; R219Q/S262N/D86V; R219Q/S262N/D86W; R219Q/S262N/D86Y; R219Q/S262N/E179C; R219Q/S262N/E179F; R219Q/S262N/E179H; R219Q/S262N/E179I; R219Q/S262N/E179K; R219Q/S262N/E179L; R219Q/S262N/E179M; R219Q/S262N/E179N; R219Q/S262N/E179P; R219Q/S262N/E179Q; R219Q/S262N/E179R; R219Q/S262N/E179W; R219Q/S262N/E179Y; R219C/S262N; R219D/S262N; R219E/S262N; R219F/S262N; R219G/S262N; R219H/S262N; R219I/S262N; R219L/S262N; R219M/S262N; R219P/S262N; R219S/S262N; R219T/S262N; R219V/S262N; R219W/S262N; R219Y/S262N; R219Q/S262N/L221C; R219Q/S262N/L221D; R219Q/S262N/L221E; R219Q/S262N/L221F; R219Q/S262N/L221H; R219Q/S262N/L221I; R219Q/S262N/L221K; R219Q/S262N/L221M; R219Q/S262N/L221N; R219Q/S262N/L221P; R219Q/S262N/L221Q; R219Q/S262N/L221R; R219Q/S262N/L221S; R219Q/S262N/L221T; R219Q/S262N/L221W; R219Q/S262N/L221Y; R219Q/S262C; R219Q/S262D; R219Q/S262E; R219Q/S262F; R219Q/S262G; R219Q/S262H; R219Q/S262I; R219Q/S262K; R219Q/S262L; R219Q/S262P; R219Q/S262Q; R219Q/S262R; R219Q/S262T; R219Q/S262W; R219Q/S262Y; R219Q/S262N/H264C; R219Q/S262N/H264D; R219Q/S262N/H264E; R219Q/S262N/H264F; R219Q/S262N/H264I; R219Q/S262N/H264K; R219Q/S262N/H264L; R219Q/S262N/H264M; R219Q/S262N/H264P; R219Q/S262N/H264R; R219Q/S262N/H264S; R219Q/S262N/H264T; R219Q/S262N/H264V; R219Q/S262N/H264W; R219Q/S262N/H264Y; R219Q/S262N/S266A; R219Q/S262N/S266C; R219Q/S262N/S266D; R219Q/S262N/S266E; R219Q/S262N/S266F; R219Q/S262N/S266G; R219Q/S262N/S266H; R219Q/S262N/S266I; R219Q/S262N/S266K; R219Q/S262N/S266L; R219Q/S262N/S266M; R219Q/S262N/S266N;

R219Q/S262N/S266P; R219Q/S262N/S266Q; R219Q/S262N/S266R; R219Q/S262N/S266T; R219Q/S262N/S266V; R219Q/S262N/S266W; R219Q/S262N/S266Y; R219Q/S262N/K267A; R219Q/S262N/K267C; R219Q/S262N/K267D; R219Q/S262N/K267E; R219Q/S262N/K267F; R219Q/S262N/K267G; R219Q/S262N/K267H; R219Q/S262N/K267I; R219Q/S262N/K267L; R219Q/S262N/K267M; R219Q/S262N/K267N; R219Q/S262N/K267P; R219Q/S262N/K267Q; R219Q/S262N/K267R; R219Q/S262N/K267S; R219Q/S262N/K267T; R219Q/S262N/K267V; R219Q/S262N/K267W; R219Q/S262N/K267Y; R219Q/S262N/V296A; R219Q/S262N/V296C; R219Q/S262N/V296D; R219Q/S262N/V296E; R219Q/S262N/V296F; R219Q/S262N/V296G; R219Q/S262N/V296H; R219Q/S262N/V296I; R219Q/S262N/V296K; R219Q/S262N/V296L; R219Q/S262N/V296M; R219Q/S262N/V296N; R219Q/S262N/V296P; R219Q/S262N/V296Q; R219Q/S262N/V296R; R219Q/S262N/V296S; R219Q/S262N/V296T; R219Q/S262N/V296W; R219Q/S262N/V296Y; R219Q/K11A/R20A; R219Q/K11A/R20A/K371A; R219Q/R20A/K371A; R219Q/K11A/K371A; S262N/K11A/R20A; S262N/K11A/R20A/K371A; S262N/R20A/K371A; and S262N/K11A/K371A, with reference to SEQ ID NO:5.

30. The variant ADA2 protein of any of claims 1-23, wherein the variant ADA2 protein comprises one or more amino acid replacement(s) selected from among replacement(s) corresponding to R219A, R219C, R219D, R219E, R219F, R219G, R219H, R219I, R219K, R219L, R219M, R219N, R219P, R219Q, R219S, R219T, R219V, R219W, R219Y, S262A, S262C, S262D, S262E, S262F, S262G, S262H, S262I, S262K, S262L, S262M, S262N, S262P, S262Q, S262R, S262T, S262V, S262W, and S262Ywith reference to amino acid positions set forth in SEQ ID NO:5.

31. The variant ADA2 protein or catalytically active portion thereof of any of claims 1-23, wherein the variant ADA2 protein comprises one or more amino acid replacement(s) selected from from among replacements corresponding R219K, R219Q, R219N, R219A,S262A, S262V, S262M, S262N, S262C, S262D, S262E, S262F, S262G, S262H, S262I, S262K, S262L, S262P, S262Q, S262R, S262T, S262W, and S262Y.

32. The variant ADA2 protein or catalytically active portion thereof of any of claims 1-31, wherein the ADA2 protein is PEGylated.

33. The variant ADA2 protein or catalytically active portion thereof of claim 19, wherein the sequence of the ADA2 portion is set forth in SEQ ID NO:273.

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

35. A catalytically active portion of the variant ADA2 protein or catalytically active portion thereof of any of claims 1-34, wherein the catalytically active portion comprises the ADA domain.

36 A variant ADA2 multimer, comprising a plurality of variant ADA2 proteins or catalytically active portions thereof of any of claims 1-35, wherein the variant ADA2 protein or catalytically active portion thereof are the same or different.

37 A variant ADA2 dimer, comprising a variant ADA2 protein or catalytically active portion thereof of any of claims 1-36.

38. A variant ADA2 dimer, comprising a variant ADA2 protein or catalytically active portion thereof of claim 19.

39. The variant ADA2 dimer or catalytically active portion thereof of claim 37 that is a homodimer comprising two variant ADA2 protein or catalytically active portions thereof that are the same.

40. The variant ADA2 dimer or catalytically active portion thereof of claim 37 that is a heterodimer comprising two variant ADA2 protein or catalytically active portions thereof that are different from each.

41. A conjugate, comprising an ADA2 protein or catalytically active portion thereof of claim 19, linked directly or indirectly via a linker to a heterologous moiety, wherein the heterologous moiety is a half-life extending moiety.

42. A conjugate, comprising an ADA2 protein or catalytically active portion thereof of any of claims 1-40, linked directly or indirectly via a linker to a heterologous moiety, wherein the heterologous moiety is a half-life extending moiety.

43. The conjugate of claim 41 or claim 42, wherein the a half-life extending moiety is a PEG and the ADA2 protein or catalytically active portion thereof is PEGylated.

44. A nucleic acid encoding a variant ADA2 protein or catalytically active portion thereof of any of claims 1-40.

45. A vector, comprising the nucleic acid molecule of claim 44.

46. An isolated cell or a cell culture, comprising the vector of claim 45.

47. The cell or cell culture of claim 46, wherein the cell is a selected from a tumor-infiltrating lymphocyte (TIL), a cytotoxic T lymphocyte (CTL), a natural killer (NK) cell or a lymphokine-activated killer (LAK) cell.

48. The cell or cell culture of claim 47, wherein the cell is a T cell that encodes and expresses chimeric antigen receptor (CAR) and the variant ADA2 protein or dimer, wherein the CAR is specific for a tumor cell antigen.

49. A method of producing an ADA2 variant or catalytically active portion thereof, comprising culturing a cell of claim 47 or claim 48, under conditions whereby the ADA2 variant is expressed.

50. The method of claim 49, wherein the resulting ADA2 variant is a dimer.

51. A method, comprising, culturing or expanding the cells of any one of claims 46-48 to produce expanded cells.

52. The expanded cells produced by the method of claim 51.

53. Use of the expanded cells of claim 52 for manufacture of a medicament treatment of a tumor or cancer.

54. The expanded cells of claim 52 for treatment of a tumor or cancer.

55. A pharmaceutical composition, comprising a variant ADA2 protein of any of claims 1-40 or a catalytically active portion thereof, or a dimer or multimer of the ADA2 protein or catalytically active portion thereof or a conjugate of any of claims 41-43.

56. The pharmaceutical composition of claim 55, wherein the ADA2 variant or catalytically active portion thereof is PEGylated.

57. The pharmaceutical composition of claim 55, wherein the ADA2 variant comprises the sequence of amino acid residues set forth in SEQ ID NO:273 or a catalytically active portion thereof.

58. The pharmaceutical composition of claim 55, wherein the ADA2 protein or catalytically active portion thereof is PEGylated.

59. An ADA variant or multimer thereof of any of claims 1-40 or the conjugate of any of claims 41-43 for treatment of a tumor, cancer, non-cancer hyperproliferative disease, a fibrotic disease, an infectious disease, a vasculopathy, or an immunodeficiency disease.

60. The use of claim 59, wherein the ADA2 protein comprises the sequence of amino acid residues set forth in SEQ ID NO:273 or a catalytically active portion thereof.

61. The use of claim 60, wherein the ADA2 protein or a catalytically active portion thereof is PEGylated.

62. Use of pharmaceutical composition of any of claims 55-58 for manufacture of a medicament treatment of a tumor, cancer, non-cancer hyperproliferative disease, a fibrotic disease, an infectious disease, a vasculopathy, or an immunodeficiency disease in a subject.

63 The use of any of claims 54 and 62-65, wherein the ADA2 protein comprises the sequence of amino acid residues set forth in SEQ ID NO:273 or a catalytically active portion thereof.

64. The use of claim 63 wherein the ADA2 protein or a catalytically active portion thereof is PEGylated.