

Richter et al. (2013, PLoS One 8:e72156) describe the inhibition of TNFR1 to interact with its natural ligands TNF and lymphotoxin alpha (LT α) by ATROSAB as measured by the release of IL-6 and IL-8 from HeLa and HT1080 cells, respectively, induced by TNF or LT α .

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SUMMARY

It was the objective to provide an improved anti-huTNFR1 agent with improved TNFR1-inhibiting characteristics while avoiding any side effects caused by intrinsic TNF mimetic agonistic activity.

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The object is solved by the subject matter as claimed.

In one aspect of the present disclosure, there is provided an inhibitor of the human tumor necrosis factor receptor 1 (huTNFR1) receptor which is a human or humanized antibody construct that monovalently recognizes huTNFR1 through an antigen-binding moiety, wherein the antigen-binding moiety comprises

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- a heavy chain variable (VH) domain that comprises the complementarity determining regions (CDR) sequences CDRH1, CDRH2, and CDRH3, and
- a light chain variable (VL) domain that comprises the CDR sequences CDRL1, CDRL2, and CDRL3, wherein:

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- a) the CDRH1 sequence is identified as SEQ ID NO: 1;
- b) the CDRH2 sequence is identified as SEQ ID NO: 10;
- c) the CDRH3 sequence is identified as SEQ ID NO: 3;
- d) the CDRL1 sequence is identified as SEQ ID NO: 4;
- e) the CDRL2 sequence is identified as SEQ ID NO: 5; and
- f) the CDRL3 sequence is identified as SEQ ID NO: 11.

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In another aspect of the present disclosure, there is provided a pharmaceutical preparation comprising the inhibitor disclosed herein and a pharmaceutically acceptable carrier.

In a further aspect of the present disclosure, there is provided a method of producing an inhibitor disclosed herein employing a recombinant mammalian expression system to express the antibody construct.

In another aspect of the present disclosure, there is provided an inhibitor disclosed herein, for use in medical use where anti-tumor necrosis factor (TNF) therapies or non-biologic disease-modifying anti-rheumatic drugs (DMARD) are indicated.

In a further aspect of the present disclosure, there is provided a use of an inhibitor disclosed herein in the manufacture of a medicament for treating a human subject in need of an anti-TNF therapy.

In another aspect of the present disclosure, there is provided an isolated nucleic acid encoding the inhibitor disclosed herein.

In a further aspect of the present disclosure, there is provided an expression vector comprising the nucleic acid disclosed herein.

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In another aspect of the present disclosure, there is provided an isolated recombinant host cell comprising the nucleic acid disclosed herein.

According to the present disclosure there is provided an inhibitor of the TNF -
5 huTNFR1 receptor interaction which is a human or humanized antibody construct that monovalently recognizes huTNFR1 through an antigen-binding moiety.

Specifically, the antigen-binding moiety comprises

- a heavy chain variable (VH) domain that comprises the CDR sequences CDRH1, CDRH2, and CDRH2, and
- 10 - a light chain variable (VL) domain that comprises the CDR sequences CDRL1, CDRL2, and CDRL2, wherein

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- a) the CDRH1 sequence is identified as SEQ ID 1;
- b) the CDRH2 sequence is identified as SEQ ID 10;
- 15 c) the CDRH3 sequence is identified as SEQ ID 3;
- d) the CDRL1 sequence is identified as SEQ ID 4;
- e) the CDRL2 sequence is identified as SEQ ID 5; and
- f) the CDRL3 sequence is identified as SEQ ID 11;

or

20 B

- a) the CDRH1 sequence is a functionally active CDR variant of SEQ ID 1;
- and/or

- b) the CDRH2 sequence is a functionally active CDR variant of SEQ ID 10;
- and/or

- 25 c) the CDRH3 sequence is a functionally active CDR variant of SEQ ID 3;
- and/or

CLAIMS

1. An inhibitor of the human tumor necrosis factor receptor 1 (huTNFR1) receptor which is a human or humanized antibody construct that monovalently
5 recognizes huTNFR1 through an antigen-binding moiety, wherein the antigen-binding moiety comprises
- a heavy chain variable (VH) domain that comprises the complementarity-determining regions (CDR) sequences CDRH1, CDRH2, and CDRH3, and
 - a light chain variable (VL) domain that comprises the CDR sequences CDRL1,
10 CDRL2, and CDRL3, wherein:
- a) the CDRH1 sequence is identified as SEQ ID NO: 1;
 - b) the CDRH2 sequence is identified as SEQ ID NO: 10;
 - c) the CDRH3 sequence is identified as SEQ ID NO: 3;
 - 15 d) the CDRL1 sequence is identified as SEQ ID NO: 4;
 - e) the CDRL2 sequence is identified as SEQ ID NO: 5; and
 - f) the CDRL3 sequence is identified as SEQ ID NO: 11.

2. The inhibitor of claim 1, wherein the antibody construct comprises an antibody selected from the group consisting of Fab molecules, scFv molecules, disulfide-stabilized Fv (dsFv), half-IgG1 antibodies, and Fv domains.

5 3. The inhibitor of claim 2, wherein the antibody construct is any of a Fab, scFv, dsFv, or Fv domains, which is fused to an antibody Fc fragment.

10 4. The inhibitor of claim 1, wherein the antibody construct is modified by linkage to polyethylene glycol (PEGylation), glycosylated PEG, hydroxyl ethyl starch (HESylation), or poly-sialic acids (PSA).

5. The inhibitor of claim 1, wherein the VH domain is an affinity matured variant of a parent VH domain identified as SEQ ID NO: 12; and the VL domain is an affinity-matured variant of a parent VL domain identified as SEQ ID NO: 16.

6. The inhibitor of claim 1, wherein

a) the VH domain comprises or consists of SEQ ID NO:13, or a functionally active variant thereof comprising at least 95% sequence identity to SEQ ID NO: 13; and/or

5 b) the VL domain comprises or consists of a sequence selected from the group consisting of SEQ ID NO: 17 or SEQ ID NO: 19, or a functionally active variant thereof comprising at least 95% sequence identity to SEQ ID NO: 17 or SEQ ID NO: 19.

7. The inhibitor of claim 1,

10 comprising a combination of a VH and a VL domain, which is

selected from the group consisting of group members i) to ii), wherein

i)

VH comprises or consists of SEQ ID NO: 13, and

15 VL comprises or consists of SEQ ID NO: 17;

and

ii)

VH comprises or consists of SEQ ID NO: 13, and
VL comprises or consists of SEQ ID NO: 19.

8. The inhibitor of claim 1, wherein the antibody construct has a thermostability of at
5 least 60°C, as determined by dynamic light scattering.

9. A pharmaceutical preparation comprising the inhibitor of claim 1 and a
pharmaceutically acceptable carrier.

10. A method of producing an inhibitor of claim 1 employing a recombinant mammalian expression system to express the antibody construct.

5 11. The inhibitor of claim 1, for use in medical use where anti- tumor necrosis factor (TNF) therapies or non-biologic disease-modifying anti-rheumatic drugs (DMARD) are indicated.

10 12. The inhibitor for use according to claim 11, wherein the subject has developed anti-drug antibodies.

13. The inhibitor for use according to claim 11, wherein the subject is suffering from
a) acute or chronic inflammation of joints, skin and gut; and/or
b) autoimmune diseases, rheumatoid arthritis, psoriasis, psoriatic arthritis, juvenile arthritis, ankylosing spondylitis, Crohn's disease, multiple sclerosis, congestive heart failure,
15 metabolic disease, cytokine release syndrome, septic shock, acute and chronic neurodegenerative disease, stroke, Alzheimer's and Parkinson's disease, colitis ulcerosa, pancreatitis, chronic obstructive pulmonary disease (COPD), acute fulminant viral or bacterial infections, chronic neurodegenerative diseases, genetically inherited diseases with TNF/TNFR1 as the causative pathologic mediator, periodic fever syndrome, cherubism, and
20 cancer.

14. Use of an inhibitor of claim 1 in the manufacture of a medicament for treating a human subject in need of an anti-TNF therapy.

25 15. The use of claim 14, wherein treatment is "first line" treatment, or "second line" treatment where anti-TNF or non-biologic DMARD therapeutics failed.

16. The use of claim 14, wherein the subject has developed anti-drug antibodies.

17. The use of any of claim 14, wherein the subject is suffering from

a) acute or chronic inflammation of joints, skin and gut; and/or

b) autoimmune diseases, rheumatoid arthritis, psoriasis, psoriatic arthritis, juvenile arthritis, ankylosing spondylitis, Crohn's disease, multiple sclerosis, congestive heart failure, metabolic disease, cytokine release syndrome, septic shock, acute and chronic neurodegenerative disease, stroke, Alzheimer's and Parkinson's disease, colitis ulcerosa, pancreatitis, COPD, acute fulminant viral or bacterial infections, chronic neurodegenerative diseases, genetically inherited diseases with TNF/TNFR1 as the causative pathologic mediator, periodic fever syndrome, cherubism, and cancer.

18. An isolated nucleic acid encoding the inhibitor of claim 1.

19. An expression vector comprising the nucleic acid of claim 18.

20. An isolated recombinant host cell comprising the nucleic acid of claim 18.

21. The pharmaceutical preparation of claim 9, wherein the preparation is formulated for parenteral use.

22. The pharmaceutical preparation of claim 21, wherein the parenteral use is by intravenous or subcutaneous administration.

23. The method of claim 10, wherein a Chinese Hamster Ovary (CHO) production cell line is employed.

24. The inhibitor for use according to claim 11, wherein treatment is "first-line" treatment, or "second-line" treatment where anti-TNF or non-biologic DMARD therapeutics failed.