PATENT COOPERATION TREATY

From the
INTERNATIONAL SEARCHING AUTHORITY

To:

HALSTEAD David, P.
Patent Group
FOLEY HOAG LLP
155 Seaport Boulevard
Boston, MA 02210-2600
United States of America

PCT

WRITTEN OPINION OF THE
INTERNATIONAL SEARCHING AUTHORITY

(PCT Rule 43*bis*.1)

Date of mailing *(day/month/year)*	27 Jul 2020
Applicant's or agent's file reference ABH-00725	**FOR FURTHER ACTION** See paragraph 2 below

International application No.	International filing date *(day/month/year)*	Priority date *(day/month/year)*
PCT/US2020/030435	29 Apr 2020	30 Apr 2019

International Patent Classification (IPC) or both national classification and IPC
IPC (20200101) C07K 16/28 C07K 16/46 C12N 5/0783 A61K 35/28 A61K 39/395 A61P 35/02
CPC (20130101) C07K 16/28 C07K 16/2896 C07K 16/46 C12N 5/0636 A61K 35/28 A61K 39/39558 A61P 35/02

Applicant
ATARA BIOTHERAPEUTICS, INC.

1. This opinion contains indications relating to the following items:

- [X] Box No. I Basis of the opinion
- [ ] Box No. II Priority
- [X] Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
- [ ] Box No. IV Lack of unity of invention
- [X] Box No. V Reasoned statement under Rule 43*bis*.1(a)(i) with regard to novelty, inventive step and industrial applicability; citations and explanations supporting such statement
- [ ] Box No. VI Certain documents cited
- [ ] Box No. VII Certain defects in the international application
- [X] Box No. VIII Certain observations on the international application

2. **FURTHER ACTION**

If a demand for international preliminary examination is made, this opinion will be considered to be a written opinion of the International Preliminary Examining Authority ("IPEA") except that this does not apply where the applicant chooses an Authority other than this one to be the IPEA and the chosen IPEA has notified the International Bureau under Rule 66.1*bis*(b) that written opinions of this International Searching Authority will not be so considered.

If this opinion is, as provided above, considered to be a written opinion of the IPEA, the applicant is invited to submit to the IPEA a written reply together, where appropriate, with amendments, before the expiration of 3 months from the date of mailing of Form PCT/ISA/220 or before the expiration of 22 months from the priority date, whichever expires later.

For further options, see Form PCT/ISA/220.

Name and mailing address of the ISA:	Date of completion of this opinion	Authorized officer
Israel Patent Office Technology Park, Bldg.5, Malcha, Jerusalem, 9695101, Israel Email address: pctoffice@justice.gov.il	26 Jul 2020	PACE Umberto Telephone No. 972-73-3927148

WRITTEN OPINION OF THE INTERNATIONAL SEARCHING AUTHORITY

International application No.
PCT/US2020/030435

Box No. I Basis of this opinion

1. With regard to the **language**, this opinion has been established on the basis of:

   [X] the international application in the language in which it was filed.

   [ ] a translation of the international application into _____ which is the language of a translation furnished for the purposes of international search (Rules 12.3(a) and 23.1(b)).

2. [ ] This opinion has been established taking into account the **rectification of an obvious mistake** authorized by or notified to this Authority under Rule 91 (Rule 43*bis*.1(b))

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, this opinion has been established on the basis of a sequence listing:

   a. [ ] forming part of the international application as filed:

      [ ] in the form of an Annex C/ST.25 text file.

      [ ] on paper or in the form of an image file.

   b. [ ] furnished together with the international application under PCT Rule 13*ter*.1(a) for the purposes of international search only in the form of an Annex C/ST.25 text file.

   c. [X] furnished subsequent to the international filing date for the purposes of international search only:

      [X] in the form of an Annex C/ST.25 text file (Rule 13*ter*.1(a)).

      [ ] on paper or in the form of an image file (Rule 13*ter*.1(b) and Administrative Instructions, Section 713).

4. [ ] In addition, in the case that more than one version or copy of a sequence listing has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that forming part of the application as filed or does not go beyond the application as filed, as appropriate, were furnished.

5. Additional comments:

WRITTEN OPINION OF THE INTERNATIONAL SEARCHING AUTHORITY

International application No.
PCT/US2020/030435

Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

The questions whether the claimed invention appears to be novel, to involve an inventive step (to be non obvious), or to be industrially applicable have not been examined in respect of:

☐ the entire international application.

☒ claims Nos. 66-82

because:

☒ the said international application, or the said claims Nos. 66-82 relate to the following subject matter which does not require an international search *(specify)*:

Claims 66-82 relate to subject matter considered by this Authority to be covered by the provisions of Rule 39.1(iv) PCT. Consequently, no opinion will be formulated with respect to the industrial applicability of the subject-matter of this claim (Article 17(2)(a)(i) PCT).

☐ the description, claims or drawings *(indicate particular elements below)* or said claims Nos. _____ are so unclear that no meaningful opinion could be formed *(specify)*:

☐ the claims, or said claims Nos. _____ are so inadequately supported by the description that no meaningful opinion could be formed *(specify)*:

☐ no international search report has been established for said claims Nos. _____

☐ a meaningful opinion could not be formed without the sequence listing; the applicant did not, within the prescribed time limit:

☐ furnish a sequence listing in the form of an Annex C/ST.25 text file, and such listing was not available to the International Searching Authority in a form and manner acceptable to it; or the sequence listing furnished did not comply with the standard provided for in Annex C of the Administrative Instructions.

☐ furnish a sequence listing on paper or in the form of an image file complying with the standard provided for in Annex C of the Administrative Instructions, and such listing was not available to the International Searching Authority in a form and manner acceptable to it; or the sequence listing furnished did not comply with the standard provided for in Annex C of the Administrative Instructions.

☐ pay the required late furnishing fee for the furnishing of a sequence listing in response to an invitation under Rule 13*ter*.1(a) or (b).

☐ See Supplemental Box for further details.

WRITTEN OPINION OF THE INTERNATIONAL SEARCHING AUTHORITY

International application No.
PCT/US2020/030435

Box No. V **Reasoned statement under Rule 43*bis*.1(a)(i) with regard to novelty, inventive step and industrial applicability; citations and explanations supporting such statement**

1. Statement

Novelty (N)	Claims	5,13-15,18-27,38-65,68,73-82	YES
	Claims	1-4,6-12,16,17,28-37,66,67,69-72	NO
Inventive step (IS)	Claims		YES
	Claims	1-82	NO
Industrial applicability (IA)	Claims	1-65	YES
	Claims		NO

2. Citations and explanations:

Relevant literature

D1: Kochenderfer, James N., et al. "B-cell depletion and remissions of malignancy along with cytokine-associated toxicity in a clinical trial of anti-CD19 chimeric-antigen-receptor - transduced T cells." Blood, The Journal of the American Society of Hematology 119.12 (2012): 2709-2720.
https://ashpublications.org/blood/article/119/12/2709/29857/B-cell-depletion-and-remissions-of-malignancy
This document teaches a T cell genetically modified to express an anti CD19 chimeric antigen receptor (CAR). The CAR taught by the document comprises the variable regions of a murine anti -human-CD19 Ab, a portion of the CD28 molecule, and the signaling domain of the CD3-zeta molecule. The document describes a clinical trial of the CAR-expressing cell for the treatment of B-cell malignancies (see the whole document).

D2: Kochenderfer, James N., et al. "Construction and pre-clinical evaluation of an anti-CD19 chimeric antigen receptor." Journal of immunotherapy (Hagerstown, Md.: 1997) 32.7 (2009): 689.
https://journals.lww.com/immunotherapy-journal/Abstract/2009/09000/Construction_and_Preclinical_Evaluation_of_an.2.aspx
This document teaches an anti CD19 CAR consists of an anti-CD19 scFv that was derived from the FMC63 mouse hybridoma 39, a portion of the human CD28 molecule, and the intracellular component of the human TCR-CD3-zeta molecule (see materials and methods).

D3: US2016362472 (A1)
This document teaches CARs and CAR-T cells directed against CD19. Sequence 111, comprising a CD19-binding region is identical to SEQ ID NO. 9 of the present application). The document teaches also a CAR comprising a 4-1BB costimulatory domain (Figure 1).

D4: Boucher, Justin C., et al. "Mutation of the CD28 costimulatory domain confers decreased CAR T cell exhaustion." Blood 132.Supplement 1 (2018): 966-966.
https://ashpublications.org/blood/article/132/Supplement%201/966/266122/Mutation-of-the-CD28-Costimulatory-Domain-Confers
This document teaches that incorporating null mutations of the CD28 subdomains (YMNM, PRRP, or PYAP) optimizes CAR T cell signaling and reduces exhaustion (see the whole document).

D5: Boucher, Justin C., et al. "Mutation of the CD28 costimulatory domain confers increased CAR T cell persistence and decreased exhaustion." (2018): 57-28.
https://www.jimmunol.org/content/200/1_Supplement/57.28.abstract
This document teaches that mutating the CD28 subdomains YMNM and PRRP and leaving only the PYAP CD28 subdomain active can optimize CAR T cell signaling and reduce exhaustion (the whole document).

D6: Love, Paul E., and Sandra M. Hayes. "ITAM-mediated signaling by the T-cell antigen receptor." Cold Spring Harbor perspectives in biology 2.6 (2010): a002485.
https://cshperspectives.cshlp.org/content/2/6/a002485.short
This document reviews the function of the ITAM regions in the signaling of the T-cell receptor. In particular, the document teaches mutants of the ITAM regions (Table 2).

D7: Qin, Le, et al. "Incorporation of a hinge domain improves the expansion of chimeric antigen receptor T cells." Journal of hematology & oncology 10.1 (2017): 1-11.
https://jhoonline.biomedcentral.com/articles/10.1186/s13045-017-0437-8
This document teaches an anti CD-19 CAR comprising a hinge domain. The CAR includes a CD-19 binding region derived from an anti-CD19 scFv, the transmembrane and intracellular domains of CD28 and the intracellular domain of CD3-zeta. Additionally, the construct comprises a hinge domain derived from IgG4.
The document teaches that incorporation of a hinge domain improves the expansion of CAR- T cells (see the whole document, especially page 2, second paragraph of the right column, Fig. 1 and the "Results" section).

D8: FMC63-28Z receptor protein [synthetic construct]
GenBank: ADM64594.1
https://www.ncbi.nlm.nih.gov/protein/ADM64594.1?report=genbank&log$=protalign&blast_rank=1&RID=HAN7S91A014
This document teaches the sequence of a chimeric antigen receptor directed against CD19.

D9: Pule, M. A., Savoldo, B., Myers, G. D., Rossig, C., Russell, H. V., Dotti, G., ... & Yvon, E. (2008). Virus-specific T cells engineered to coexpress tumor-specific receptors: persistence and antitumor activity in individuals with neuroblastoma. Nature medicine, 14(11), 1264.
https://www.nature.com/articles/nm.1882
This document teaches the preparation of virus-specific T cells engineered to express CARs. Virus specific CTL's were prepared by stimulation of PBMCs with gamma irradiated autologous LCLs. And subsequently transformed with retroviral supernatant encoding a chimeric antigen receptor directed against the diasialoganglioside GD2, a tumor -associated antigen expressed by human neuroblastoma cells.
The CTLs were used for treating individuals with relapsed neuroblastoma (see the whole document).

D10: WO 2017/027291
This document teaches a bi-specific T-cell comprising two CARs. The cells are produced by simultaneous transduction with lentiviruses expressing an anti CD19 and an anti CD20 CAR (paragraphs 32-33 and figures 3 and 4). Also shown are CAR T-cells expressing chimeric receptors against CD19 and CE7 (paragraph 39, figure 9), CD19 and ROR1 (paragraph 46 and figure 11). Other tumor antigens are considered, such as glypican-3 (GPC3), NY-ESO-1, SSX-2 (paragraph 11).

D11: Rafiq, Sarwish, et al. "Targeted delivery of a PD-1-blocking scFv by CAR-T cells enhances anti-tumor efficacy in vivo." Nature biotechnology 36.9 (2018): 847-856.
https://www.nature.com/articles/nbt.4195
This document teaches a method of improving the efficacy of a CAR T cell therapy by using CAR T cells further modified to express a PD-1 blocking antibody (see the whole document).
.

D12: Zhang, Yongping, et al. "CRISPR-Cas9 mediated LAG-3 disruption in CAR-T cells." Frontiers of medicine 11.4 (2017): 554-562.
https://link.springer.com/article/10.1007/s11684-017-0543-6
This document teaches a method to improve efficacy of a CAR T cell by knocking out the negative regulator LAG-3. The knockout is done by CRISPR-Cas9-mediated gene editing (see the whole document).

D13: Yoon, Dok Hyun, et al. "Incorporation of immune checkpoint blockade into chimeric antigen receptor T cells (CAR-Ts): combination or built-in CAR-T." International journal of molecular sciences 19.2 (2018): 340.
https://www.mdpi.com/1422-0067/19/2/340
This document teaches the addition of immune checkpoint blockade to a CAR T cell therapy by all the different possible routes: expression of dominant negative receptors or a switch receptor, knockdown/knockout of immune checkpoint molecule, expression of inhibitory molecules and combination therapy (see the whole document).

Novelty

The present application does not meet the requirements of Article 33(1) PCT since claims 1-4, 6-12, 16-17, 28-37, 66-67 and 69-72 lack novelty in the sense of Article 33(2) PCT.

Documents D1 and D2 teach a chimeric antigen receptor (CAR) directed against CD19. The CAR comprises a CD19 binding region, an scFv derived from a monoclonal antibody, a portion of the CD28 molecule, including the transmembrane region and a costimulatory domain, and the signaling domain of CD3-zeta. Also the use of 4-1BB costimulatory domain is described.
In the light of the teaching of D1 and D2 claims 1-4, 6-12, 16-17 lack novelty. The CAR of D1 and D2 was expressed in T cells, from a viral vector and therefore also claims 28-37 lack novelty.
D1 and D2 teach the clinical use of the engineered CAR-T cells for the treatment of B cell malignancies. Therefore, claims 66-67 lack novelty as well.
Since the treatment was done on patients that underwent chemotherapy also claims 69-72 are not novel.

Inventive step

The present application does not meet the requirements of Article 33(1) PCT since claims 1-82 lack an inventive step in the sense of Article 33(3) PCT.

Claims 1-4, 6-12, 16-17, 28-37, 66-67 and 69-72 are not novel and therefore are not inventive

as well.

The remaining claims comprise additional features. These features are not specifically detailed in D1 and D2 but they are taught by the additional cited prior art, in the same context of chimeric antigen receptor-expressing T cells. Therefore the claims are not inventive due to the combined teaching of D1/D2 with the prior art document D3-D13, as detailed below.

Claim 5,26 relates to the sequence of the CD19 binding domain (SEQ ID NO. 9). A CD19 binding region comprising this sequence is not explicitly mentioned by D1 or D2 but it is taught by D3.

Claims 13-15 and 56-58 relate to CAR molecules comprising mutations of the CD28 signaling domain in the subdomains YMNM, PRRRP and PYAP. These mutations, as means of increasing CAR T cell persistence and decreasing exhaustion are taught by D4 and D5.

Claims 18-21 relate to a mutant CD3-zeta comprising a mutated or deleted ITAM (immunoreceptor tyrosine-based activation motif). Mutations of ITAM (tyr to phe mutations) are known from the prior art (see D6, Table 2 and references thereof).

Claims 22-23, 59 relate to the addition of a hinge sequence to the CAR. This feature is known from D7. The use of a hinge from other molecules of the immunoglobulin superfamily is an obvious choice.

Claims 24-25 relate to the addition of a leader sequence. This is a well-known feature in the preparation of recombinant molecules and does not provide an inventive step.

Claims 24-25 relate to the addition of a signal peptide. This feature is known from general knowledge in the field and does not provide an inventive step.

Claim 27 relates to the full sequence of a CAR polypeptide of the invention. The structure of this CAR has been described (see D8). The sequence in D8 is almost identical to sequence ID NO. 7, except for the mutations in the ITAM regions, which were known from D6 and do not provide an inventive step (see above).

Claims 38-41 relate to the use of a CTL as therapeutic CAR-containing cell, particularly CTLs that have been sensitized by exposition to a virus. D9 teaches the preparation of virus (EBV)-sensitized CTLs, engineered to express CARs.

Claims 42-55 relate to bi-specific CAR T cells, which express two CARs, one of which recognizes a B-lymphocyte antigen (e.g. CD19) and the other one is directed against a tumor antigen. D10 teaches bi-specific CAR T cells. The claims recite features of CARs that were already mentioned in earlier claims: claims 45-46 are parallel to claims 3-4, claim 47 is parallel to claim 8, claims 48-52 are parallel to claims 17-20, claim 53-55 are parallel to claim 10-12. All this claims lack an inventive step due to the combined teachings of D10 with the previously mentioned citations.

Claims 60-65 relate to CAR T cells modified to prevent expression of immune checkpoint molecules (ICM), or expressing a dominant-negative form of ICM or expressing a switch receptor for a ICM or expressing antibodies that block ICM signaling. D11-D13 teach methods

of combining CAR T cell therapy with the blockage, by various means, of immune checkpoint inhibition. D13 reviews all the technology and is relevant to all the claims. D11 teaches the expression of a PD-1 blocking antibody and is relevant to claims 63-65. D12 teaches the knock-out of LAG-3 and is relevant to claims 60-61.

Claim 68 relates to the treatment of an EBV-associated lymphoproliferative disorder. D9 teaches the preparation of a CAR T cell which is responsive to an EBV antigen.

Claims 73-74 relate to the source of the T-lymphocytes used for the CAR-T cell preparation. These are standard features that do not provide an inventive step.

Claims 75-82, relate to a combined therapy with CAR T cells and an immune checkpoint inhibitor- externally given or expressed by the CAR T cell. All these additional features are taught by D12-D13.

Industrial Applicability

The subject-matter of claims 1-65 are considered to be industrially applicable in the sense of Article 33(4) PCT. For the assessment of the present claims 66-82 on the question whether it is industrially applicable, no unified criteria exist in the PCT Contracting States. The patentability can also be dependent upon the formulation of the claims. The ILPO, for example, does not recognize as industrially applicable the subject matter of claims relating to a method of medical treatment of the human body, but may allow claims relating to a compound for use in the medical treatment of the human body.

WRITTEN OPINION OF THE INTERNATIONAL SEARCHING AUTHORITY

International application No.
PCT/US2020/030435

Box No. VIII Certain observations on the international application

The following observations on the clarity of the claims, description, and drawings or on the question whether the claims are fully supported by the description, are made:

Claim 1 does not comply with the requirements of Article 6 PCT since the subject matter is presennted in general terms. The scope of the claim is so wide that cannot be meaningfully searched. The search and examination of claim 1 is based on the features of the dependent claims.