

PATENT COOPERATION TREATY

From the
INTERNATIONAL SEARCHING AUTHORITY

To: see form PCT/ISA/220	PCT WRITTEN OPINION OF THE INTERNATIONAL SEARCHING AUTHORITY (PCT Rule 43bis.1)	
Date of mailing (day/month/year) see form PCT/ISA/210 (second sheet)		
Applicant's or agent's file reference see form PCT/ISA/220	FOR FURTHER ACTION See paragraph 2 below	
International application No. PCT/US2022/074575	International filing date (day/month/year) 05.08.2022	Priority date (day/month/year) 05.08.2021
International Patent Classification (IPC) or both national classification and IPC INV. C07K16/28		
Applicant ELI LILLY AND COMPANY		

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

- ☒ Box No. I Basis of the opinion
- ☐ Box No. II Priority
- ☐ Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
- ☐ Box No. IV Lack of unity of invention
- ☒ Box No. V Reasoned statement under Rule 43bis.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
- ☒ Box No. VIII Certain observations on the international application

2. FURTHER ACTION

If a demand for international preliminary examination is made, this opinion will usually 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 66.1bis(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:  European Patent Office P.B. 5818 Patentlaan 2 NL-2280 HV Rijswijk - Pays Bas Tel. +31 70 340 - 2040 Fax: +31 70 340 - 3016	Date of completion of this opinion see form PCT/ISA/210	Authorized Officer Hix, Rebecca Telephone No. +31 70 340-0
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Box No. I Basis of the opinion

1. With regard to the **language**, this opinion has been established on the basis of:
 - ☒ 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 43bis.1(a))
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.
 - b. ☐ furnished subsequent to the international filing date for the purposes of international search (Rule 13ter.1(a)),
 - ☐ accompanied by a statement to the effect that the sequence listing does not go beyond the disclosure in the international application as filed.
4. ☐ With regard to any nucleotide and/or amino acid sequence disclosed in the international application, this opinion has been established to the extent that a meaningful opinion could be formed without a WIPO Standard ST.26 compliant sequence listing.
5. Additional comments:

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

1. Statement

Novelty (N)	Yes: Claims	<u>10-14, 18-35, 41, 47, 52</u>
	No: Claims	<u>1-9, 15-17, 36-40, 42-46, 48-51</u>
Inventive step (IS)	Yes: Claims	<u>10-14, 21-24, 30</u>
	No: Claims	<u>1-9, 15-20, 25-29, 31-52</u>
Industrial applicability (IA)	Yes: Claims	<u>1-52</u>
	No: Claims	

2. Citations and explanations

see separate sheet

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:

see separate sheet

Item V

Reasoned statement under Rule 43bis1(a)(i) with regard to novelty, inventive step or industrial applicability.

- 1 For the assessment of the present claims involving methods of treatments; Claims 37 to 40 relate to subject-matter considered by this Authority to be covered by the provisions of Rule 39.1(iv) / 67.1(iv) PCT. The patentability can be dependent upon the formulation of the claims. The EPO, for example, does not recognise as patentable claims to the use of a compound in medical treatment, but may allow claims to a product, in particular substances or compositions for use in a first or further medical treatment.
- 2 The following **documents** are referred to in this communication;
 - D1 WO 2020/048312 A1 (SUNSHINE GUOJIAN PHARMACEUTICAL SHANGHAI CO LTD [CN]) 12 March 2020 (2020-03-12)
 - D2 US 2013/336978 A1 (ERIKSSON PER-OLOF FREDRIK [SE] ET AL) 19 December 2013 (2013-12-19)
 - D3 KIM JUNG-EUN ET AL: "Engineering of anti-human interleukin-4 receptor alpha antibodies with potent antagonistic activity",
SCIENTIFIC REPORTS,
vol. 9, no. 1, 1 December 2019 (2019-12-01), XP055858581,
DOI: 10.1038/s41598-019-44253-9
Retrieved from the Internet:
URL: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6533264/pdf/41598_2019_Article_44253.pdf
 - D4 D M PATON: "Dupilumab: human monoclonal antibody against IL-4Ralpha for moderate to severe atopic dermatitis",
DRUGS TODAY,
vol. 53, no. 9, 1 September 2017 (2017-09-01), pages 477-487, XP055465888,
DOI: 10.1358/dot.2017.53.9.2693150

D5 AUDREY LE FLOC'H ET AL: "Dual blockade of IL-4 and IL-13 with dupilumab, an IL-4R[alpha] antibody, is required to broadly inhibit type 2 inflammation", ALLERGY, WILEY-BLACKWELL PUBLISHING LTD, UNITED KINGDOM, vol. 75, no. 5, 3 January 2020 (2020-01-03), pages 1188-1204, XP071463316, ISSN: 0105-4538, DOI: 10.1111/ALL.14151

3 NOVELTY (Art. 33(2) PCT)

- 3.1 D1 discloses antibodies targeting IL-4R α as alternatives to the known anti-4R α duplimab. D1 describes the construction of the chimeric antibody 4-2-Chimeric-IgG1, humanized antibodies 4-2-Humanized-IgG1 and 4-2-Humanized-IgG4. The experimental results show that the chimeric antibody 4-2-Chimeric-IgG1 and the humanized antibodies 4-2-Humanized-IgG1 and 4-2-Humanized-IgG4 can effectively block the interaction between IL-4 and IL-4R and inhibit IL-4 induced TF-1 cell proliferation and human peripheral blood mononuclear cells (PBMC) expression of CD23 and secretion of IgE. The antibody developed in D1 is used to prepare a therapeutic drug for patients with IL-4R overexpression diseases, including atopic dermatitis, asthma, allergic reactions, eosinophilic esophagitis, skin infections, and nasal polyposis, see claim 18.
- 3.2 Example 8 Epitope Analysis of Anti-human IL-4R Humanized Antibody where analysis of the antibody epitope showed that the antibodies specifically bind to the following amino acid residues on the extracellular domain of IL-4R α : L39, F41, L42, L43, D72, and Y74. Table 2 shows the binding effects of 4-2-Hu-IgG4 and Dupilumab on IL-4R α mutants, showing that the epitope comprises D66, D67 and D125. Figure 6, shows that Dupilumab, 4-2-Chimeric-IgG1, and 4-2-Humanized-IgG1 can effectively inhibit IL-13-induced proliferation of TF-1 cells, with IC50 of 73.8ng / ml, 81.6ng / ml, and 34.3ng / ml.
- 3.3 In the light of the disclosures of D1, the subject-matter of claims 1 to 9, 15 to 17, 36 to 40, 42 to 46 and 48 to 51 is not new according to Article 33(2) PCT.
- 3.4 D2 discloses an antibody or fragment thereof that specifically binds human interleukin-4 receptor alpha (hIL-4R α). Table 5, page 37 shows that the anti-hIL-4R α antibodies binding epitopes including L64, M65, R99. Example 3.2 describes the inhibition of IL-13 and IL-4 Induced Proliferation of TF-1 Cells by IgG.
- 3.5 In the light of the disclosures of D1, the subject-matter of claims 1 to 3, 8 and 9 is not new according to Article 33(2) PCT.

- 3.6 D3 isolated anti-human IL-4R α antagonistic Abs and engineered their complementarity determining regions to improve the affinity to generate a candidate Ab, 4R34.1.19. When reformatted as human IgG1 form, 4R34.1.19 specifically bound to IL4R α with a high affinity ($K_D \approx 17$ B pM) and effectively blocked IL-4- and IL-13-dependent signaling in a reporter cell system at a comparable level to that of the clinically approved anti-IL-4R α dupilumab Ab analogue. Epitope mapping by alanine scanning mutagenesis revealed that 4R34.1.19 mainly bound to IL-4 binding sites on IL-4R α with different epitopes from those of dupilumab analogue.
- 3.7 Figure 4, page 6 shows that the IL-4 analogue, hIL-4-mFc, significantly lost the binding to IL-4R mutants carrying an alanine substitution at Y38, D92, V93, D97, and Y208 (Fig. 4c). The isolated anti-IL-4R α Abs substantially lost the binding activities with considerable varying degrees for the alanine mutants at L67, L68, D92, V93, and D97 residues (Fig. 4c, Supplementary Fig. 4c), indicating that they mainly recognize the ligand binding regions on IL-4 R α but their epitopes are slightly different to each other.
- 3.8 D4 is a review of the anti-human IL-4R α antibody Dupilumab and its use in treating severe to moderate atopic dermatitis.
- 3.9 D5 describes Dupilumab, a fully human monoclonal antibody that binds IL-4R α and inhibits signaling of both IL-4 and IL-13, has shown efficacy across multiple diseases with underlying type 2 signatures and is approved for treatment of asthma, atopic dermatitis, and chronic sinusitis with nasal polyposis.
- 3.10 In the light of the prior art cited, the subject-matter of claims 1 to 9, 15 to 17, 36 to 40, 42 to 46 and 48 to 51 is not new according to Article 33(2) PCT.
- 3.11 In view of the prior art cited, claims 10 to 14, 18 to 35, 41, 47 and 52 appear to be novel and meet therefore the requirements of Art. 33(2) PCT, since none of the anti-human IL-4R α antibodies disclosed in the prior art comprise the amino acid sequences of the six CDRs or VH or VL sequences according to independent claims 10, 12, 21, 25 or 30 or where the anti-human IL-4R α antibody has been used to treat cancer according to claims 41, 47 and 52.

4 INVENTIVE STEP (Art. 33(3) PCT)

- 4.1 Document D1 is considered to represent the most relevant state of the art.

- 4.2 In the light of the above difference, the effect of said difference is shown as the following in the description:
- Example 2 determines the structural and functional epitope of the human IL-4R α Antibodies;
 - Example 2a involves the Structural epitope determination of the 8660 Fab by X-ray crystallography which show that the 8660 Fab antibody epitope overlapped with both the IL-4 and the IL-13 binding sites to IL-4R, thus indicating that binding of the **exemplified antibodies to IL-4R α would physically block the IL-4 and IL-13 cytokines from binding to IL-4R α when the Fab variant portion of the exemplified antibodies is bound to IL-4R α .**
 - Example 2b involves the functional epitope determination of the 5F3 IgG4PAA.
 - Table 3 show that the functional epitope for the exemplified 5E3 IgG4PAA antibody comprises amino acid residues D66 and D125. Substitution of amino acid residue D125 to Arginine, which is located near amino acid residue D66 on the crystal structure of the IL-4R α (see Figure 2), also showed significantly reduced binding of 0.59 OD₄₅₀.
 - Table 3a shows the functional epitope determination of exemplified human IL-4R α antibodies.
 - Example 2c determines the structural Epitope of the Human IL-4R α 5559 Antibodies where the 5559 Fab antibody epitope overlapped with both the IL-4 and the IL-13 binding sites to IL-4R α , indicating that binding of the exemplified 5559 antibodies would physically block the IL-4 and IL-13 cytokines from binding to IL-4R α .
 - Example 2d determines the functional epitope of the human IL-4R α 5559 Antibodies.
 - Table 3b shows the functional epitopes of the human IL-4R α 5559 antibodies.
 - Example 3 investigates the binding affinity and functional activity of the human IL-4R α Antibodies, the results are shown in;
 - Table 4, and Figures 3A and 3B, show that the exemplified 5559 anti-IL-4R α antibody variants had a significant increase in binding affinity to both human and cynomolgus monkey IL4-R α when compared to the 5F3 IgG4PAA antibody.
 - Table 5A, and Figures 4A and 4B, show that the exemplified IL-4R α antibodies 5559 IgG1 A 124C/378C and 5559 IgG4P KRQ 124C/378C bound with comparable affinity to the human PBMC isolated B cells (EC₅₀ of 0.14 nM and

0.15 nM, respectively) and CD4 T cells (EC_{50} of 28.7 nM and 26.3 nM, respectively). The results showed that the KRQ amino acid residue substitutions and the 124C/378C amino acid residue substitutions did not impact binding of the exemplified anti-IL-4R α 5559 antibodies to the B or T cells.

- Table 5B and Figure 4C show that the 5559 IgG1 AAA 124C/378C effector null antibody showed unexpectedly reduced affinity to B cells (EC_{50} of 1.07 nM) when compared to the 5559 IgG1 A 124C/378C antibody (EC_{50} of 0.27 nM), indicating that the Fc portion of the antibody may impact binding of the exemplified IL4R α antibody to B cells.

- Table 5A shows the binding of exemplified human IL-4R α antibodies to B and T cells.

- Table 6 and Figures 5A and 5B, show that the exemplified IL-4R α antibodies **5559 IgG1A 124C/378C, 5559 IgG4P KRQ 124C/378C and 8660 IgG4P 124C/378C inhibited both IL-4 (Figure 5A) and IL-13 (Figure 5B) induced SEAP activity in a dose dependent manner.**

- Table 7, and Figures 6A and 6B show that the exemplified IL-4R α antibodies **inhibited IL-4 induced STAT6 phosphorylation in both CD4⁺ T cells (Figure 6A) and B cells (Figure 6B) in a dose dependent manner.**

- Table 8 and Figure 6C show that the exemplified IL-4R α antibodies also **inhibited IL-13 induced STAT6 phosphorylation in B cells (Figure 6C) in a dose dependent manner.**

- Table 9 and Figure 7 show that the exemplified IL-4R α antibodies inhibited IL-4 induced B cell proliferation in a dose dependent manner.

- Table 10 and Figures 8A and 8B show that the exemplified IL-4R α antibodies inhibited both IL-4 (Figure 8A) and IL-13 (Figure 8B) induced CD23 expression on myeloid cells.

- Example 4 investigates the effector function activity of the human IL-4R α Antibodies.

- Table 11 summarizes the binding affinities (K_D) of the exemplified IL-4R α antibodies 5559 IgG4P KRQ 124C/378C and 5559 IgG1A 124C/378C to human Fc γ R1, Fc γ RIIa, Fc γ RIIb, and Fc γ RIIIa receptor ECDs.

- Figure 9 shows that the exemplified antibodies 5559 IgG1A 124C/378C, and 5559 IgG4P KRQ 124C/378C did not bind complement component C1q.

- Figure 10A shows that the exemplified IL-4R α antibodies 5559 IgG1A 124C/378C, 5559 IgG4P KRQ 124C/378C and 5559 IgG4P 124C/378C significantly lacked or had no ADCC activity, when compared to the positive control.
 - Figure 11 shows that the exemplified IL-4R α antibodies 5559 IgG1A 124C/378C, 5559 IgG4P KRQ 124C/378C, and 5559 IgG4P 124C/378C did not induce CDC activity when compared to the positive control.
 - Example 5 and Table 12 show the Biophysical Properties of the Human IL-4R α Antibodies.
- 4.3 The problem to be solved by the present invention may therefore be regarded as providing alternative anti-human IL-4R α antibodies.
- 4.4 The proposed solution are anti-human IL-4R α antibodies that bind to the extracellular domain (ECD) of IL-4R α as provided by SEQ ID NO: 15. Tables 1 and 2, pages 30 and 31 provide the CDR and amino acid sequences of the anti-human IL-4R α antibodies of the present application;
- Anti-human IL-4R α antibody 5559 comprises CDRs SEQ ID NOs 1 to 6 , VH/VL SEQ ID NOs 7/8 according to claims 10 and 11.
 - Anti-human IL-4R α antibody 8660 comprises CDRs SEQ ID NOs 42, 2, 3, 22, 5 and 6 , VH/VL SEQ ID NOs 44/45 according to claims 12 to 13.
 - Anti-human IL-4R α antibody 5F3 IgG4PAA comprises CDRs SEQ ID NOs 19, 20, 3, 22, 5 and 24 , HC/LC SEQ ID NOs 27/28, VH/VL SEQ ID NOs 25/26 according to claims 21 to 23.
- 4.5 Although D1 discloses anti-human IL-4R α humanized antibodies that specifically binds to the following amino acid residues on the extracellular domain of IL-4R α : L39, F41, L42, L43, D72, and Y74 and effectively block the interaction between IL-4 and IL-4R and inhibit IL-4 induced TF-1 cell proliferation and human peripheral blood mononuclear cells (PBMC) expression of CD23 and secretion of IgE and effectively inhibit IL-13-induced proliferation of TF-1 cells, there is no indication in any of the available prior art that the particular amino acids in the ECD would result in anti-human IL-4R α antibodies that also inhibit binding of IL-13 and IL-4 to human IL-4R α .
- 4.6 Consequently the subject-matter of independent claims 10, 12, 21 and 30, and dependent claims 11, 13, 14 and 22 to 24, which define the anti-human IL-4R α antibodies disclosed in the present application in terms of comprising the amino

acid sequences of the six CDRs or the VH **and** VL and the composition according to claim 30, are considered to involve an inventive step according to Article 33(3) PCT.

- 4.7 However independent claim 25 defines a nucleic acid encoding one of the sequences of one of either the VH or VL sequences of the antibodies of the application. The same applies to claim 29, where the problem has only been demonstrated to be solved by the anti-human IL-4R α antibodies disclosed in the present application comprising the amino acid sequences of the six CDRs or the VH **and** VL.
- 4.8 Consequently the subject-matter of claims 25 to 29 and claims dependent thereon; claims 31 to 35 are not considered to solve the technical problem in an inventive manner as they do not provide the solution to the problem according to Article 33(3) PCT.
- 4.9 As far as claims 41, 47 and 52 are concerned, directed to the medical use of the anti-human IL-4R α antibodies to treat cancer, there is no indication in the present application that the antibodies plausibly may be used to treat cancer, particularly since the anti-human IL-4R α antibodies of the prior art are directed to the treatment of IL-4R associated disorders such as immune inflammatory disorders as listed in claim 46.
- 4.10 Therefore there is no indication that the subject-matter of claims 41, 47 and 52 provide any inventive solution and are therefore not considered to involve an inventive step contrary to Article 33(3) PCT.
- 4.11 In view of the above, the present application does not meet the requirements of Article 33(3) PCT, because the subject-matter of claims 15 to 20, 25 to 29 and 31 to 52 does not involve an inventive step.
- 4.12 In view of the above, the present application meets the requirements of Article 33(3) PCT, because the subject-matter of claims 10 to 14 and 21 to 24 involves an inventive step in the sense of Article 33(3) PCT.

Item VIII

Certain observations on the international application

5 CLARITY (Art.6 PCT)

- 5.1 Since the three CDRs of each of the variable domains of the light and heavy chains are normally responsible for binding to the antigen, the conventional antibody, in order to be uniquely defined by its structure only and have its characteristic binding specificity, needs to be defined by at least these six CDRs or by the amino acid sequences of the VH and the VL, in order to fulfil the requirements of Art. 6 PCT. As independent claims 1, 25 and 29 do not clearly and unambiguously define the anti-human IL-4R α antibodies of the present application in such terms, said claims are considered to unclear according to Article 6 PCT.
- 5.2 The term "the epitope comprises one or more amino acid residues selected from ..." used in claim 1 is vague and indefinite and as such renders the scope of the claims unclear, according to Art. 6 PCT, since the epitope to which the anti-human IL-4R α antibodies of the present application should be defined in terms of the particular amino acids which are bound by the antibodies.
- 5.3 Claims 41, 47 and 52 are directed to the medical use of the anti-human IL-4R α antibodies to treat cancer, where there is neither any support or disclosure of the use of the anti-human IL-4R α antibodies of the present application used to treat cancer in the description or examples. As such the subject-matter of claims 41, 47 and 52 are not supported by the description according to Article 6 PCT.