

OPINION UNDER SECTION 74A

Patent	EP 1257290 B2
Proprietor(s)	Florian Kern
Exclusive Licensee	
Requester	Greaves Brewster LLP
Observer(s)	
Date Opinion issued	22 January 2019

The request

1. The comptroller has been requested by Greaves Brewster LLP (“the Requester”) to issue an opinion on whether certain claims of EP 1257290 B2 (“the Patent”) are valid on the grounds of lacking novelty or an inventive step in light of seven documents, D1-D7, supplied by the Requestor.
2. The request was received on 22 October 2018. The request was accompanied by a statement explaining the request as well as copies of the cited documents.
3. Although the representatives (Ladas & Perry LLP) of the patent proprietor Florian Kern (“the Proprietor”) requested and were allowed an extension to file observations, there were no observations nor consequently any observations in reply.
4. The Patent entitled ‘Method for antigen-specific stimulation of T-lymphocytes with synthetic peptides’ was filed on 17 February 2001 under the provisions of the Patent Cooperation Treaty (PCT) with international application number PCT/EP01701773 in the name of Florian Kern. The application claimed an earliest priority date of 22 February 2000 and was initially published as WO 01/63286. After entering the European regional phase, the Patent was granted on 7 January 2009 in amended form following opposition proceedings at the EPO and remains in force in the UK. As the Requester states this patent is in German, but there are translated granted claims in both French and English in addition to German in the final B2 specification. Further the Requester identifies an English language equivalent US patent, US 7994096 B2, and asserts it has an identical description and drawings to the patent in question, although the scope of the claims in the US granted patent was restricted to human cytomegalovirus antigen-specific T lymphocyte stimulation. Where it is necessary, I will therefore refer to the relevant sections of this US patent.

5. The documents supplied by the Requestor representing the prior art were all published before the filing and priority dates of the patent, and so may be considered as prior art for the purposes of novelty or inventive step, or as representing the Common General Knowledge (CGK). These documents are listed below using the same numbering used by the Requester:

D1: Protti et al, J Immunology, Vol 144, No 4, 1990, pages 1276-1281

D2: Protti et al, J Immunology, Vol 144, No 5, 1990, pages 1711-1720

D3: Bixler et al, Immunology, Vol 56, 1985, pages 103-112

D4: Kern et al, Nature Medicine, Vol 4, No 8, 1998, pages 975-978

D5: Raju et al, Vol 9, No 1, 1996, pages 79-88

D6: Reece et al, J Immunol Methods, Vol 172, 1994, pages 241-254

D7: Kern et al, J Virology, 1999, pages 8179-8184

Whether all parts of the request are allowable

6. The comptroller will not issue an opinion if for any reason he considers it inappropriate in all the circumstances to do so (by virtue of section 74A(3)(b) of the Patents Act 1977 – “the Act”). In particular, requests will be refused which do no more than repeat arguments already considered pre-grant. Here, the Requester requests an opinion on whether certain claims of the Patent are invalid on the basis of lack of inventive step with respect to document D4. I note that D7, which is prior art from the Proprietor, has been listed by the Requestor as representing the common general knowledge (CGK) of the skilled person in their consideration of D4. However as the Requester notes, D7 was disclosed in the description of the patent as forming part of the background to the invention, and also document D4 was identified in the search report as a category A citation. The Requester also states that D4 was further mentioned during the examination procedure and in the opposition proceedings in respect of novelty and in a Notice of Opposition filed by a third party company.
7. In light of this, I consider that the relevance of this document was considered pre-grant and during the opposition process of the EPO. I therefore refuse the Request to consider whether this document also raises an inventive step objection. I will however, consider the questions of novelty and inventive step raised by the Requester in respect of the other documents they have cited, D1-D3 and D5-D6 that have not been raised previously.
8. In their concluding remarks the Requester refers to claims 15 and 17 as lacking novelty or an inventive step. However, no arguments or analysis of the documents cited are presented concerning these claims and so I have not considered these claims.

The Patent

9. The Patent relates to a method for antigen-specific stimulation of T-lymphocytes with synthetic peptides which can identify those cells that respond to and/or the peptides that cause the stimulation, as well as the strength of the stimulation. The Patent explains that the prior art does not allow either determining systemically in a single measurement whether a T lymphocyte response against a target protein is present at all nor how strong this response is (see column 2, lines 10-15), whereas in the invention of the Patent protein antigens of a known sequence are employed to immunostimulate CD8⁺ and CD4⁺ T lymphocytes without the need to identify individual epitopes or for the protein to undergo cellular antigen processing. As a result, it can be established whether an organism from which the T lymphocytes have been obtained has built up a response after exposure to the immunising antigen, and a population of such cells can be propagated.

Claim 1

10. The patent contains claims 1-17 of which claim 1 is the sole independent claim. Claims 2-12 set out further specific aspects of the method. Claims 13-17 further relate to compositions and stimulating mixtures obtainable or identified by the method. I shall begin by considering claim 1. It will only be necessary for me to consider those claims dependent on claim 1 that the Requestor argues also lack novelty or an inventive step if I find that claim 1 lacks novelty. Claim 1 defines the invention in the following terms:

1. A method for the antigen-specific stimulation of CD8⁺ and/or CD4⁺ T lymphocytes with synthetic peptide libraries in vitro that is suitable for detecting a T cell immune response or for preparing a CD8⁺ and/or CD4⁺ T lymphocyte composition for the in vivo treatment of humans and animals, comprising the following steps:

(a) subdividing the amino acid sequence of the total protein into protein fragments with partial amino acid sequences, wherein said protein fragments have a minimum length of 9 amino acid residues (AAs) and a maximum length of 25 AAs, and wherein adjacent or neighboring protein fragments are overlapping with their partial amino acid sequence;

(b) synthesizing a peptide library containing the protein fragments defined in (a); and

(c) incubating a suspension containing the CD8⁺ and/or CD4⁺ T lymphocytes to be stimulated with all the protein fragments of said peptide library in a single culture run

Construction of claim 1

11. To consider the validity of the claims of the Patent, I first need to construe them. That is to say I must interpret them in the light of the description and drawings as instructed by Section 125(1) of the Act. In doing so I must interpret the claims in context through the eyes of the person skilled in the art. Ultimately the question is

what the person skilled in the art would have understood the patentee to be using the language of the claims to mean.

12. The Requester has broken down each of steps (a)-(c) of the method of claim 1 into a series of distinct features thus:

Step (a):

(a1) subdividing the amino acid sequence of the total protein into protein fragments with partial amino acid sequences,

(a2) wherein said protein fragments have a minimum length of 9 amino acid residues (AAs) and a maximum length of 25 AAs, and wherein

(a3) adjacent or neighboring protein fragments are overlapping with their partial amino acid sequence;

Step (b):

(b) synthesizing a peptide library containing the protein fragments defined in (a); and

Step (c):

(c1) incubating

(c2) a suspension containing the CD8+ and/or CD4+ T lymphocytes to be stimulated with all the protein fragments of said peptide library in a single culture run

13. In part 8, section 8.1 of their request, “*Applying the Windsurfer/Pozzoli approach favoured by the UKIPO, ...*” the Requester has construed the invention of claim 1 not to be any different from the method for antigen-specific stimulation of T-lymphocytes with synthetic peptides as set out in the claim, and that is suitable for detecting a T cell immune response or for preparing a CD8⁺ and/or CD4⁺ T lymphocyte composition for the in vivo treatment of humans and animals. Where necessary in their request, the Requester provides their interpretation of each of features set out in the steps of the method in their analysis of the documents they have cited. However, many of these are straightforward to construe for the person skilled in the art and require no further comment
14. As such I see no need to construe the scope of the claim any differently from the approach taken by the requester, nor do I need to consider the meaning of the features identified above further because these would be clear to a person skilled in the art.
15. I consider the person skilled in the art to be an immunologist, as identified by the Requester. I thus agree with the Requester.

Novelty and Inventive step – the law

16. The relevant provisions in relation to novelty and inventive step are Sections 1-3 of the Act.
17. Section 1(1)(a) and (b) of the Act reads:

1(1) A patent may be granted only for an invention in respect of which the

following conditions are satisfied, that is to say

(a) the invention is new;

(b) it involves an inventive step;

18. The relevant provisions in relation to novelty are found in section 2(1) and section 2(2) which read:

2(1) An invention shall be taken to be new if it does not form part of the state of the art.

2(2) The state of the art in the case of an invention shall be taken to comprise all matter (whether a product, a process, information about either, or anything else) which has at any time before the priority date of that invention been made available to the public (whether in the United Kingdom or elsewhere) by written or oral description, by use or in any other way.

19. The provisions in relation to inventive step are found in section 3 which states:

3. An invention shall be taken to involve an inventive step if it is not obvious to a person skilled in the art, having regard to any matter which forms part of the state of the art by virtue only of section 2(2) above (and disregarding section 2(3) above).

Whether claim 1 lacks novelty in light of the cited prior art

20. The Requester submits that claim 1 lacks novelty when compared to any one of four documents D1-D3 and D5.

21. I will consider the Requestor's arguments in respect of D1 first.

22. D1 is an article titled 'CD4⁺ T cell response to human acetylcholine receptor a subunit in myasthenia gravis: A study with synthetic peptides' by Protti et al which was published in The Journal of Immunology in 1990. The human acetylcholine receptor (AcChR) was subdivided into a series of overlapping fragments and these were then used in a microproliferation assay with a population of CD4⁺ enriched T cells to determine an anti-AcChR response of the cells. The Requester argues that each of features (a1)-(c2) of claim 1 is disclosed by D1. I shall consider each feature in turn with respect to the disclosures in D1.

Feature (a1)

23. I agree with the Requester that D1 discloses the generation a pool of 32 peptides corresponding to the AcChR protein and this is equivalent to subdividing the total protein amino acid sequence into protein fragments with partial amino acid sequences as required by feature (a1).

Feature (a2)

24. The 32 peptides produced in D1 each have a length of 14 to 20 amino acids and I agree with the Requester that feature (a2) wherein the peptide fragments can be

between 9 and 25 amino acids residues long is disclosed as required.

Feature (a3)

25. Feature (a3) requires that the partial peptide fragments overlap with adjacent or neighbouring peptides. The peptides of D1 overlap each other and so I agree that feature (a3) is also disclosed.

Feature (b)

26. The Requester directs me to passages in the Material and Methods of D1 disclosing the synthesis of the partial peptides spanning the entire AcChR protein by manual parallel synthesis. I agree with the Requester that this synthesis represents the synthesis of a "peptide library" of overlapping fragments defined in (a) as required by feature (b) of the claim.

Feature (c1)

27. The Requester argues that the term "incubating" of this feature would be understood by the skilled person to be within the scope of the "microproliferation assay" by which the pool of peptides are used to stimulate CD4⁺ enriched cells. I agree that this would be the understanding of the term "incubating" as required in feature c1.

Feature (c2)

28. The microproliferation assay described in the Materials and Methods of D1 applies different concentrations of what is identified as an " α pool of peptide fragments" that span the complete AcChR peptide to a population of Cd4⁺ T cells in a single well of a microtitre plate to stimulate said cells. I therefore agree with the Requester that this feature is also disclosed in D1.
29. From the above analysis I agree with Requester that the invention as defined in claim 1 is disclosed by the method set out in D1. The disclosure must also be an 'enabling disclosure'. In other words the skilled person must be able to work the disclosed invention, using trial and error experiments if necessary. In my view, given the disclosure of each feature identified above and experimental protocols to carry each one out, it would be straightforward for the skilled person to use their expertise and knowledge to carry out the invention defined in claim 1. Therefore in my opinion claim 1 is not novel in light of D1.
30. The Requester has similarly asserted that each of D2, D3 and D5 demonstrates that claim 1 lacks novelty over the disclosures in these citations. The requester has applied the same feature based approach to identify the disclosures in each of these documents that relate to the relevant feature.
31. D2 is an academic publication titled "Use of synthetic peptides to establish anti-human acetylcholine receptor CD4⁺ cell lines from myasthenia gravis patients" from the same authors as D1 and which was also published in The Journal of Immunology in 1990. The Materials and Methods of this document disclose the use of the same α pool of peptides spanning the entire length of the AcChR protein in the same microproliferation assays with CD4⁺ T cells as disclosed in D1. Consequently, I see

no need for me to repeat the in depth feature analysis set out above, but I agree with the Requester that D2 also discloses all the features of claim 1 and so it is not novel over D2 as well.

32. D3 is a further academic citation by Bixler et al and was published in 1985. It is entitled "Antigen presentation of lysozyme: T-cell recognition of peptide and intact protein after priming with synthetic overlapping peptides comprising the entire protein chain". Consecutive overlapping peptides spanning the chicken lysozyme protein were prepared, used to induce an immune response in mice and then lymph cells harvested from these mice were challenged with these peptides to measure their proliferative responses. The Requester has asserted that this document discloses all the features of claim 1. I will therefore consider the Requester's arguments.

Feature (a1)

33. D3 discloses that the lysozyme protein has been subdivided into a series of protein fragments and so I agree with the Requestor that the feature of (a1) is disclosed.

34. *Feature (a2)*

35. The synthetic lysozyme fragments disclosed in this document are between 19 and 21 residues in length. I agree with the requester that this feature is also disclosed.

Feature (a3)

36. I agree with the Requester that the fragments are disclosed as being overlapping as required by feature (a3).

Feature (b)

37. I further agree with the Requester that the set of overlapping peptides that span the entire lysozyme protein form a library as required by this feature.

38. *Features (c1, c2)*

39. The Requester asserts that the Challenge experiment detailed in Table 3 of D3 where the challenge is performed with the peptide mixture against lymph node cells in a proliferation assay (which I note is similar to those disclosed in documents D1 and D2) and which comprises an equimolar mixture of all of the peptides spanning lysozyme, represents the two features (c1) and (c2). Whilst D3 does not specifically identify the markers present on the T lymphocytes that are isolated from the immunised mice and used in the assay, antigen-naïve T cells expand and differentiate into memory and effector T cells after they encounter their cognate antigen. These memory T cells may be either CD4⁺ or CD8⁺. As the Requestor notes, lymph nodes are disclosed in the patent that this review is concerned with as one source of T lymphocytes. Consequently, I consider that the cells being challenged in D3 include CD4⁺ and/or CD8⁺ T cells and so this is an implicit teaching of this document.

40. I therefore agree with the Requester that claim 1 is not novel over this document as it discloses all the features of claim 1.

41. The Requester has finally asserted that claim 1 lacks novelty over document D5. This academic article was published in the Journal of Autoimmunity by Raju et al in 1996 and is entitled "Epitope repertoire of Human CD4⁺ lines propagated with tetanus toxoid or synthetic tetanus toxin sequences". As the title suggests the experiments describe include challenging CD4⁺ cells with peptides spanning tetanus toxoid and assessing the response of these cells to this challenge.

Feature (a1)

42. The Requester identifies that the tetanus toxin peptide was divided into 87 overlapping synthetic peptides spanning the whole peptide, which meets the requirement of feature (a1).

Feature (a2)

43. The peptides disclosed in D5 are 20 amino acids long and I agree with the Requester that this is as required by feature (a2).

Feature (a3)

44. Furthermore as required by feature (a3) these peptides are disclosed as overlapping by 5 residues, and so I agree with the Requester on this point.

Feature (b)

45. Document D5 discloses that these peptides were synthesised by manual parallel synthesis, and were pooled to form a "peptide pool" thus comprise a complete panel of the peptides screening both the tetanus toxin H and L chains. I agree with the Requester that this forms a "library" as required by feature (b), and as this term would be understood by the skilled person.

Feature (c1)

46. I agree with the Requester that D5 discloses a step in the protocols described that encompasses the term "incubating" and so feature (c1) is fulfilled.

Feature (c2)

47. The CD4⁺ T cells are incubated with the "peptide pool" of D5 that represents the peptides spanning the complete tetanus toxin in a single experiment, as well as sub pools of these peptides and the full length peptide in separate experiments. Therefore I agree with the Requester that this feature is therefore also disclosed by D5.

48. D5 therefore discloses all the features of the invention of claim 1.

49. Consequently, I am of the opinion that claim 1 lacks novelty in light of any one of documents D1, D2, D3 or D5.

Whether claims 2, 4-6, 8-13 lack novelty in light of the cited prior art

50. I will now consider briefly the validity of the remaining claims 2, 4-6, 8-13 as requested. As explained above I consider claim 1 to lack novelty in light of documents D1, D2 D3 or D5.
51. The Requester asserts that documents D1 and D2 can be further cited against claims 2, 4, and 6, D3 can be cited against claims 6 and 11 and that D5 can be cited against claims 4-6 and 8-13. These claims are set out below:
- 2. The method according to claim 1, wherein an overlap of 8 AAs, preferably 11 AAs, exists between neighboring protein fragments.*
 - 3. The method according to one or more of claims 1 and 2, wherein the synthetic protein fragments are extended by a maximum of 7 natural or artificial AAs and/or a protective group at the N terminus and/or C terminus.*
 - 4. The method according to one or more of claims 1 to 3, wherein the concentration of the individual protein fragments of the peptide library is at least 1 ng/ml, preferably from 0.1 to 10 µg/ml, in the culture mix.*
 - 5. The method according to one or more of claims 1 to 4, wherein one or more compounds having costimulatory properties and selected from the costimulatory antibodies, such as anti-CD28 and anti-CD49d, and CTLA4-Ig are added to the incubation solution.*
 - 6. The method according to one or more of claims 1 to 5, wherein the total amino acid sequence of the total protein is determined prior to step (a) in the method.*
 - 7. ...*
 - 8. The method according to one or more of claims 1 to 4 which is capable of establishing whether T-lymphocyte-stimulating antigenic determinants are present in an antigen.*
 - 9. The method according to one or more of claims 1 to 6 which is adapted for in vitro immunostimulation of T lymphocytes of mammals, especially humans.*
 - 10. The method according to claim 9, further comprising expansion of the stimulated T lymphocytes.*
 - 11. The method according to one or more of claims 1 to 6 which is suitable for detecting a T cell immune response, namely to establish whether a mammal, especially a human, has previously responded to at least one protein fragment of the whole protein of step (a) with its immune system, and if so, how strong such response is.*
 - 12. The method according to one or more of claims 8 to 11 which comprises*

the use of several different synthetic peptide libraries, wherein the incubation of the peptide libraries with the CD8+ and/or CD4+ T lymphocyte suspension is effected together in one culture run or in separated culture runs.

13. A stimulated CD8+ and CD4+ T lymphocyte composition obtainable by the method according to claim 9 or 10.

52. Both documents D1 and D2 disclose peptides that overlap with neighbouring peptides by between 4 and 8 residues, and that these peptides are used to stimulate the cells in the microproliferation assays at concentrations of 0.05, 0.1, 0.5, 1.0 and 5 ug/ml per peptide. I agree with the Requester that either of these documents discloses the features of claims 2 and 4. I further agree with the requester that document D5 also discloses the use of the pooled peptides at a final concentration of 1 ug/ml per peptide and so also anticipates the feature of claim 4.
53. All four of documents D1, D2, D3 and D5 disclose the synthesis of the peptide fragments of the target proteins being used to stimulate the T lymphocytes. As such I agree with the Requester that this implicitly requires that the sequence of the protein is known in order to synthesise the fragments, and so claim 6 is not novel.
54. The Requester asserts that D5 discloses the use of co-stimulatory compounds in the method because the medium can contain T cell growth factor (TCGF), which is a co-stimulatory compound, and so results in claim 5 which requires that the medium contains one or more compounds having costimulatory properties lacking novelty. However, claim 5 further defines that the compounds are “...*selected from the costimulatory antibodies, such as anti-CD28 and anti-CD49d, and CTLA4-Ig ...*” TCGF does not fall into the category of a costimulatory antibody and so I do not consider that claim 5 lacks novelty over D5.
55. The requester also asserts that claims 8-13 lack novelty over document D5. I agree with the Requester that the methods disclosed in D5 are “... *capable of establishing whether T-lymphocyte-stimulating antigenic determinants are present in an antigen*” as required by claim 8, given that it discloses a dose dependent response to tetanus toxin. In addition, because the T cells are obtained from a human source, claim 9 also lacks novelty. Further claim 10, which defines the expansion of the stimulated T lymphocytes, also lacks novelty given that D5 further discloses the propagation of the T cells identified. Similarly, claim 13, which defines a cell population obtained by the methods of claims 9 or 10 also lacks novelty given the disclosures in document D5, which discloses that the T cells are human and are propagated.
56. I agree with the Requester that claim 11 lacks novelty over D5 which makes clear that the cells are obtained from humans exposed to tetanus toxoid through routine vaccination and as such have “... *previously responded to at least one protein fragment of the whole protein of step (a) with its immune system, and if so, how strong such response is.*” The method in D5 can establish how strong the T cell response is as set out in the claim.
57. D5 discloses that the tetanus toxin peptide fragments can be separated into different pools that are then used in costimulation assays and so I agree with the Requester that claim 12 is not novel in the light of this disclosure.

58. I am therefore of the opinion that claims 2, 4, 6, and 8-13 are not novel for the reasons set out above in light of documents D1, D2, D3 or D5.
59. The Requester has only produced cursory comments indicating that should I consider the claims not to be novel, then the claims lacks inventive step based on the disclosure of any one of documents D1-D3 or D5. I have found claim 1 at least to be not novel in light of any one of D1-D3 or D5. Therefore there is no need for me to consider inventive step in respect of these documents.

Whether claims 2, 3 lack an inventive step in light of the cited prior art

60. The requester also asserts that claims 2 and 3 lack an inventive step in the light of the disclosures in document D6 when it is considered in combination with any one of documents D1, D2, D3 or D5. D6 discloses investigating the effects of different overlaps of peptide fragments of a protein in peptide pools on T cell epitope screening, in this example the tetanus toxin, in particular where a 12 mer peptide can be offset by 1, 2 or 3 residues (see Table 4 for example).
61. Although I have already found that claim 2 lacks novelty when the disclosures of either of documents D1 or D2 are considered, I agree that when D6 is combined with either one of documents D3 or D5 specifying the specific peptide overlap as defined in claim 2 would be obvious to the skilled person. Such a person would be aware that overlapping adjacent peptides are known in the art, and from D6 that this may be beneficial in the methods of the invention.
62. Document D6 also discloses that blocking the ends of these peptides results in peptides that were generally more efficient as stimulatory peptides. In this respect I agree with the Requester that the skilled person would consider the teaching of this document in combination with the teaching of any one of documents D1, D2, D3 or D5 for lack of inventive step against claim 3. The inclusion of a blocking agent as described in D6, which represents the feature of including a protective group on the peptide defined in claim 3, would be obvious to the skilled person, given that these can result in more efficient stimulation in the methods of the invention.
63. Therefore I conclude that neither of claims 2 or 3 is inventive over the prior art cited.

Opinion

64. It is my opinion that the invention of the Patent as defined in claim 1 is not novel in light of any one of documents D1, D2, D3 or D5. I am also of the opinion that dependent claims 2, 4 or 6 are not novel over documents D1 or D2. Furthermore neither of claims 6 or 11 is novel over document D3. In addition claims 4, 6, and 8-13 are not novel over document D5. I also consider that claim 2 is obvious when D6 is considered in combination with either one of D3 or D5, whilst similarly claim 3 lacks an inventive step when document D6 is considered in combination with any one of D1, D2, D3 or D5.

Application for review

65. Under section 74B and rule 98, the proprietor may, within three months of the date of issue of this opinion, apply to the comptroller for a review of the opinion.

Patrick Purcell
Examiner

NOTE

This opinion is not based on the outcome of fully litigated proceedings. Rather, it is based on whatever material the persons requesting the opinion and filing observations have chosen to put before the Office.