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Datasheet for the decision of 23 May 2014

Case Number: T 0342/11 - 3.3.08

Application Number: 03749280.8

Publication Number: 1572943

IPC: C12N15/02, C12N15/00

Language of the proceedings: EN

Title of invention:
CIRCULAR NUCLEIC ACID VECTORS, AND METHODS FOR MAKING AND USING THE SAME

Applicant:
The Board of Trustees of The Leland S. Stanford Junior University

Headword:
Minicircles/BOARD OF TRUSTEES STANFORD

Relevant legal provisions:
EPC Art. 123(2), 54, 56

Keyword:
Main request - Article 123(2) EPC (no)
Auxiliary Request - Requirements of the EPC met

Decisions cited:

Catchword:

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Case Number: T 0342/11 - 3.3.08

D E C I S I O N
of Technical Board of Appeal 3.3.08
of 23 May 2014

Appellant: (Applicant)	The Board of Trustees of The Leland S. Stanford Junior University 900 Welch Road, Suite 350 Palo Alto, CA 94304 (US)
Representative:	Heaton, Joanne Marie Stevens, Hewlett & Perkins 1 St. Augustine's Place Bristol BS1 4UD (GB)
Decision under appeal:	**Decision of the Examining Division of the European Patent Office posted on 31 August 2010 refusing European patent application No. 03749280.8 pursuant to Article 97(2) EPC.**

Composition of the Board:

Chairman	M. Wieser
Members:	B. Stolz
	J. Geschwind

Summary of Facts and Submissions

I. The appeal lies against the decision of the examining division, dated 8 July 2010, to refuse European patent application No. 03 749 280.8. The examining division decided that neither the main request nor any of auxiliary requests 1 to 3 met the requirements of Articles 123(2), 83, 84 and 54 EPC.

II. With its grounds of appeal, filed on 7 January 2011 (erroneously dated 7 January 2010), the appellant filed a new main request comprising claims 1 to 21 as well as a new auxiliary request comprising claims 1 to 10.

III. The appellant was summoned to oral proceedings. A communication pursuant to Article 15(1) of the Rules of Procedure of the Boards of Appeal (RPBA) was annexed to the summons.

IV. The appellant informed the board that it would not attend the oral proceedings.

V. Oral proceedings were held on 23 Mai 2014 in the absence of the appellant.

VI. Claims 1 to 10 of the main request and the auxiliary request are identical.

Claim 1 of both requests reads:

"1. An in vitro method for preparing a circular double-stranded nucleic acid vector, the method comprising:
contacting a parent nucleic acid comprising:
(a) an expression cassette comprising a promoter operably linked to a sequence encoding a

protein and flanked by attB and attP sites recognised by a unidirectional site-specific cpC31 integrase; and

(b) a coding sequence for the unidirectional site-specific φ C31 integrase under the control of an inducible promoter;

with an inducer of the inducible promoter under conditions sufficient to produce the circular double-stranded nucleic acid vector."

Claims 2 to 10 of both requests refer to specific embodiments of the subject matter of claim 1.

VII. Claim 11 of the main request reads:

"11. A circular double stranded DNA vector produced by a method according to any one of the preceding claims, in which the vector is devoid of an origin of DNA replication and a selectable marker gene, said origin of DNA replication and selectable marker gene having been removed by recombination between two φ C31 integrase-specific substrate sequences in the parent nucleic acid according to claim 1 or 2, the vector comprising a product hybrid sequence of φ C31 integrase and said expression cassette comprising a promoter operably linked to a sequence encoding a protein, said vector being characterized in that it provides for persistent expression of said protein for at least three weeks at a level that is up to 560-fold more than that of the unrecombined parent nucleic acid."

Claims 12 to 21 of the main request refer to various kits and uses of the vector according to claim 11.

VIII. The arguments of the appellant as far as relevant for the present decision can be summarized as follows:

Main request

Article 123(2) EPC

Claim 11 corresponded to former claim 1 and was further amended to introduce *inter alia* the feature that it provided for persistent expression of said protein for at least three weeks at a level that was up to 560-fold more than that of the unrecombined parent. Basis for this amendment could be found on page 25 of the application as originally filed.

Auxiliary Request

Article 123(2) EPC

Claims 1 to 10 met the requirements of Article 123(2) EPC. Support for new claims 2 and 3 could be found on page 28, lines 20 to 21 and on page 11, lines 2 to 9.

Articles 84 EPC

The terminology found to be unclear by the examining division (sections 15.2 and 4.1 to 4.3 of the decision under appeal) was no longer present in the amended claims.

Articles 54 and 56 EPC

The examining division indicated that it considered claims 13 to 16, 18, 19 and 24 of the main request before it to be novel, and it did not raise any

inventive step objections against these claims. Claims 1 to 10 were based on these claims and hence novel and inventive over the prior art.

IX. The appellant requested that the decision under appeal be set aside and a patent be granted on the basis of the main request or the auxiliary request.

Reasons for the Decision

Admissibility

1. The main request, filed with the grounds of appeal, is based on auxiliary request 4, which was filed late in the examination procedure on 5 July 2010. The request was not admitted into examination proceedings because it was prima facie not allowable (see point 15.2 of the decision under appeal). The claims were amended to address the issues raised in points 15.2.1 to 15.2.3 of the decision under appeal.

The auxiliary request, filed with the grounds of appeal, is based on claims 1 to 9 of auxiliary request 4 with amendments to address the objection raised against claim 2 (cf. point 15.2.2 of the decision under appeal).

The amendments are straightforward and considered to be a direct response to the decision under appeal. They do not introduce new ambiguities and do not add to the complexity of the case. They are therefore admitted into the procedure.

Main request

Article 123(2) EPC

2. Claim 11 is in the format of a product-by-process claim. It refers to a circular double stranded DNA vector characterised by the presence of a product hybrid sequence of φ C31 integrase and a sequence encoding a protein under the control of a promotor element. The vector is characterised by the persistent expression of said protein for at least three weeks at a level that is up to 560-fold more than that of the unrecombined parent vector.

3. Thus, claim 11 on the one hand is characterised by structural features of a rather general nature, such as the presence of the φ C31 product hybrid sequence and a sequence encoding a protein, and on the other hand by functional features relating to the expression level of the encoded protein. Basis for the general structural features can be found in original claims 1 to 4 or on page 11, lines 17 to 28 of the international patent application as published (hereinafter referred to as the application as filed). Basis for the feature relating to a 560-fold expression can, however, only be found in the context of two very specific examples.

4. Page 25, lines 18 to 31, of the application as filed, describes the result of expression of a human α1-antitrypsin (hAAT) gene from a particular minicircle vector (comprising a specific promoter) in mouse liver. It is also stated that mice receiving this minicircle DNA "produced 10- to 13- fold more serum hAAT than those receiving the purified expression cassette, which was 200- to 560- fold higher than that of ccDNA group". Furthermore, on page 27, line 8, it is disclosed that "minicircles expressed 45- to 560-fold more serum hFIX and hAAT than their parent unrecombined plasmids in mouse liver". These are the only passages of the entire

patent application as originally filed which mention 560-fold expression levels.

There is however no direct and unambiguous disclosure, neither explicit nor implicit, of the entire subject matter of claim 11, i.e. of an expression vector characterized by a 560-fold increase in the expression level of **any** protein under **any** unspecified promoter in **any** unspecified tissue. The subject matter of claim 11 is thus the result of a combination of elements from the general description and one single feature taken from a very specific example.

According to established case law of the Boards of Appeal, this non-disclosed combination of features (intermediate generalisation) is considered to be a non-allowable amendment and contravenes the requirements of Article 123(2) EPC (see Case Law of the Boards of the EPO, 7th Ed., 2013, Chapter II.E.1.2.).

5. The main request does therefore not meet the requirements of Article 123(2) EPC.

Auxiliary request

Article 123(2) EPC

6. Claims 1 and 4 to 10 correspond to claims 16, 11 to 13, and 17 to 20, respectively, of the main request before the examining division. The examining division did not raise any objections against these claims and the board sees no reason to raise any of its own motion.

7. The subject matter of new claims 2 and 3 additionally refers to the presence of an origin of replication and a selectable marker (claim 2), in particular an

antibiotic resistance gene (claim 3). The presence of an origin of replication derives from page 19, line 30 and page 20, line 9, of the application as filed, where reference is made to bacterial plasmids suitable for replication and to plasmids that are replicated in bacteria, respectively. The presence of a selectable marker, in particular of an antibiotic resistance gene, in the vector sequence is described in the paragraph bridging pages 10 and 11 of the application as filed. Furthermore, the plasmids described in part A of the section Materials and Methods comprise an origin of replication and antibiotic resistance genes.

8. The requirements of Article 123(2) EPC are therefore met.

Articles 83 and 84 EPC

9. The claims are clear and there is no evidence that the claimed subject matter could not be put into practice by a person skilled in the art.

Article 54 EPC

10. The method of claim 1 is not directly and unambiguously derivable from the first priority application US 60/407,344 (cf. point 3 of the examining division's communication of 29 August 2007). The relevant date for the assessment of novelty and inventive step is therefore 16 April 2003, the date of the second priority application US 60/463,672. Document D5, published on 24 October 2002, is thus prior art under Article 54(2) EPC.

11. The method of claim 1 includes the use of a parent nucleic acid comprising an expression cassette encoding

a protein under the control of a promoter, flanked by recombination attB and attP sites, and an inducible sequence coding for the φ C31 integrase.

12. None of the cited prior art, including document D5, discloses the use of a parent plasmid according to claim 1. The claimed subject matter is therefore novel.

Article 56 EPC

13. The closest prior art is represented by document D5 which discloses methods for the production of minicircle vectors obtained from parent plasmids comprising an expression cassette flanked by recombination sites. The purpose of document D5 is to provide vectors without toxic/inhibitory bacterial sequences. In one embodiment, the use of bacteriophage φ C31 attB and attP sites and φ C31 integrase is disclosed (page 9, lines 9 to 20 of document D5). The φ C31 integrase gene is provided as a separate piece of nucleic acid.

14. The technical problem underlying the present invention is seen as the provision of an alternative method of producing minicircle vectors free of inhibitory bacterial sequences.

15. As a solution, the patent proposes the method of claim 1, including the use of a parent plasmid comprising a sequence encoding a gene of interest and the φ C31 integrase.

16. According to pages 24 and 25 of the application as filed, expression of parent vectors encoding either the gene of human factor IX or human α1-antitrypsin and φ C31 integrase yielded the desired minicircles. The

board is therefore satisfied that the claimed method indeed solves the underlying technical problem.

17. It remains to be established whether the claimed solution involves an inventive step.

18. The available prior art discloses the production of minicircle vectors for the expression of genes of interest using various recombination based system. Documents D1 and D4 disclose the use of a φ lambda integrase, documents D2 and D3 the use of the φ P1 derived Cre/LoxP system. Documents D6, D8 and D9 disclose various properties of the φ C31 integrase system. Document D7 is an excerpt from a textbook and relates to properties of the φ lambda integrase.

None of these documents suggests or points to the production of minicircle vectors from parent vectors comprising nucleic acid encoding both, a gene of interest **and** φ C31 integrase or any other phage derived integrase.

The claimed solution is therefore not obvious, neither on the basis of document D5 alone nor on the basis of document D5 in combination with any of the other documents on file.

19. The auxiliary request therefore meets the requirements of the EPC.

Order

For these reasons it is decided that:

1. The decision under appeal is set aside.

2. The case is remitted to the Examining division with the order to grant a patent on the basis of claims 1 to 10 of the auxiliary request filed with letter of 7 January 2011, figures 1 to 5 and the sequence listing as filed, and the description yet to be adapted

The Registrar: The Chairman:

A. Wolinski M. Wieser

Decision electronically authenticated