DECISION
of 2 December 2003

Case Number: T 0012/01 – 3.3.8
Application Number: 90907718.2
Publication Number: 0471011
IPC: C12N 15/12
Language of the proceedings: EN
Title of invention:
Human Lactoferrin cDNA Sequence
Patentee:
BAYLOR COLLEGE OF MEDICINE
Opponent:
Pharming Group NV
Headword:
Lactoferrin/BAYLOR COLLEGE OF MEDICINE
Relevant legal provisions:
EPC Art. 54, 56, 83, 87, 123(2)
EPC R. 88
Keyword:
"Main request: added matter (no)"
"Sufficiency of disclosure (yes)"
"Entitlement to priority date (yes)"
"Novelty (yes)"
"Inventive step (yes)"
Decisions cited:
G 0002/98, T 1212/97
Catchword:
Case Number: T 0012/01 - 3.3.8

Decision of the Technical Board of Appeal 3.3.8 of 2 December 2003

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Decision under appeal: Decision of the Opposition Division of the European Patent Office posted 20 October 2000 revoking European patent No. 0471011 pursuant to Article 102(1) EPC.

Composition of the Board:
Chairman: L. Galligani
Members: T. J. H. Mennessier
          C. Rennie-Smith
Summary of Facts and Submissions

I. The patentee (appellant) lodged an appeal against the decision of the opposition division, given at oral proceedings on 18 November 1999 with written reasons posted on 20 October 2000, revoking the European patent No. 0 471 011. The patent was granted on European application No. 90 907 718.2 which originated from an international application published as WO 90/13642 (to be referred to in the present decision as the application as filed). Priority was claimed from American patent application US 348270 filed on 5 May 1989.

II. Whereas two parties (opponents 1 and 2) had opposed the patent, one of them (opponent 2) withdrew its opposition on 25 March 1996 and, thereby, ceased to be a party to the opposition proceedings. Opponent 1 is the present respondent.

III. The patent had been opposed on the grounds as set forth in Articles 100(a) and (b) EPC that the invention was not new (Article 54 EPC), did not involve an inventive step (Article 56 EPC) and was not sufficiently disclosed (Article 83 EPC), and on the ground as set forth in Article 100(c) EPC that the patent contained added matter (Article 123(2) EPC).

IV. Basis for the decision under appeal were the main request filed on 22 May 1998 and auxiliary requests I to IV filed on 18 November 1999. Reasons for the revocation were lack of novelty (in view of document D3 (see paragraph XI, infra)) of claims 7 to 9 of the main request and lack of inventive step (in view of document
D37 in combination with either document D1 or documents D2 and D3 (see paragraph XI, infra) of claims 1 to 9 of auxiliary request III. Auxiliary requests I, II and IV were considered to contain amendments which had not been occasioned by a ground of opposition (Rule 57a EPC).

V. With its statement of grounds of appeal, on 28 February 2001 the appellant filed a claim request to replace all the requests then on file. That request corresponded exactly to the main request on which the decision under appeal was based.

VI. A communication under Article 11(2) of the Rules of Procedure of the Boards of Appeal presenting some preliminary and non-binding views of the board was then sent to the parties. In particular, comments were made on the issues arising under Article 123(2) EPC, namely the replacement in Figure 2 of "Gln" by "Glu", the amendments to Table 1 and the feature "derived from", and those arising under Article 54 EPC.

VII. On 31 October 2003 the appellant filed further observations accompanied by two auxiliary requests denoted "A" and "B".

VIII. In a letter of 24 November 2003 confirmed by a letter of 26 November 2003, the respondent notified the board that it would not be represented at the oral proceedings.

IX. The oral proceedings took place on 2 December 2003.
X. The main request for all designated Contracting States except ES consisted of 10 claims which read:

"1. A cDNA coding for an iron-binding protein derived from a human lactoferrin, characterized in that it comprises sequence coding for the amino acid sequence 74 to 275 of Figure 2."

"2. A cDNA according to Claim 1, characterized in that it comprises the DNA sequence 237 to 842 of Figure 2."

"3. A cDNA according to Claim 1, coding for a human lactoferrin, characterized in that it comprises sequence coding for the amino acid sequence 20 to 711 of Figure 2."

"4. A cDNA according to Claim 3, characterized in that it comprises the DNA sequence 75 to 2150 of Figure 2."

"5. A cDNA according to Claim 3, characterized in that it further comprises sequence coding for the signal peptide amino acid sequence 1 to 19 of Figure 2."

"6. A cDNA according to Claim 4, characterized in that it further comprises the DNA sequence 18 to 74 of Figure 2."

"7. A synthetic iron-binding protein derived from a human lactoferrin and produced from cDNA according to Claim 1 or 2."

"8. A synthetic human lactoferrin produced from cDNA according to Claim 3 or 4."
"9. A synthetic human lactoferrin produced from cDNA according to Claim 5 or 6."

"10. A method of producing a protein claimed in Claim 7, 8 or 9, characterized by expressing cDNA claimed in Claim 1 or 2, 3 or 4 or 5 or 6, respectively in a eucaryotic expression system."

Apart from a different spelling of the term "characterized" in claims 1 to 6 and 10, claims 1 to 10 exactly corresponded to claims 1 to 10 as granted.

During oral proceedings corresponding claims (1 to 7) for the Contracting State ES were filed. These claims corresponded exactly to claims 1 to 6 and 8 as granted for ES, save for the different spelling in claims 1 to 6 of the term "characterized".

XI. The following documents are cited in the present decision:

D1  Thomas A. Rado et al., Blood, Vol. 70, No. 4, October 1987, Pages 989 to 993;

D2  Marie-Hélène Metz-Boutigue et al., Eur. J. Biochem., Vol. 145, 1984, Pages 659 to 676;


D8  Xiping Wei et al., Blood, Vol. 72, Suppl. I, 1988, Abstract No. 530;
D31 Declaration of Dr Thomas Rado dated 11 August 1999 and annexed Exhibits A to F;

D32 Declaration of Dr Xiping Wei dated 4 June 1999 and annexed Exhibits A to D;

D34 Cover page and page 1.9 of "The 1988 CLONTECH Products and Protocols Catalogs";

D37 Undated "Product Analysis Certificate" of CLONTECH Laboratories, Inc. concerning a human breast cDNA library denoted "HL 1037a";


D49 Declaration of Dr Kathryn Stowell dated 22 October 1999.

XII. The appellant's arguments, insofar as they are relevant to the present decision, may be summarised as follows:

Main request for all designated Contracting States except ES

Article 123(2) and Rule 88 EPC

The person skilled in the art would have recognised that the invention pertained to the isolation and elucidation of a cDNA coding for human lactoferrin. As such, the cDNA was the most important aspect of the invention, and the amino acid sequence was deduced from that cDNA as indicated on page 4, lines 14 to 18 of the application as filed. Because the base triplet GAA at
the respective codon position solely and necessarily encoded the amino acid glutamic acid (Glu) and not glutamine (Gln), he or she would have immediately recognised that as the result of a clerical error the amino acid designation Gln was the wrong amino acid residue designation in position 296 in Figure 2 and that the only possible correction was the one offered: amino acid Glu.

As regards the amended Table 1 in the patent specification, it pointed to the differences in the amino acid sequence of the invention vis à vis that known from document D2 which was explicitly referred to therein. Document D2 directly and unambiguously disclosed that the amino acid positions 155, 156 and 321 (according to the numbering system of the invention) were respectively Phe, Leu and Lys. In document D2, which employed a different numbering system, the corresponding positions were 135, 136 and 300. Simple sequence alignments of short amino acid stretches of the sequence of Figure 2 of the application as filed with the sequence of document D2 therefore revealed that the amino acid residues were actually identical. Thus, reference to these residues in Table 1 as filed had been removed as they were manifestly erroneous. This correction was also obvious to the skilled person.

*Article 87 EPC*

In view of the fact that the content of the description and drawings of the priority document was essentially identical with the content of the description and drawings of the application as filed, the claimed
priority was valid, i.e. claims 1 to 10 were entitled to the priority date of 5 May 1989.

Article 83 EPC

The subject-matter of the present invention was the provision of both the entire cDNA coding for human lactoferrin and the encoded protein itself. The exact sequences of these products were disclosed in the patent specification. Thus, the specification provided a sufficiently clear and complete disclosure of the claimed invention.

Article 54 EPC

Document D3 disclosed neither of the claimed cDNAs and proteins. Thus, the claimed subject-matter was novel over that document.

There was no unambiguous description of what had been disclosed by Dr Rado at the "San Antonio Meeting" held in December 3-6, 1988 (see documents D31 and D32). The criteria as set forth in decision T 1212/97 of 14 May 2001 for an oral disclosure to be novelty-destroying were not met.

Article 56 EPC

The skilled person might have attempted to generate lactoferrin cDNA clones by combining document D1 or document D2 with document D37. This attempt might also have generated cDNA clones. Nevertheless, these clones would have had unusual properties, namely they would have had an unexpectedly low frequency, they would have
been incomplete and they would have failed to encode a protein corresponding to the established amino acid sequence data. Those properties would have entirely undermined their authenticity based on the state of the art.

This would have imposed the need to obtain independent validation of the cDNA sequences. Nevertheless, all of the obvious routes of validation were associated with intrinsic problems. Because (i) the robustness of the prior art was a disincentive against further polypeptide sequencing, (ii) additional protein sequence determination would have imposed an undue burden, (iii) the Clontech library of document D37 could not validate or complete the cDNAs obtained, (iv) no other suitable source of tissue mRNA was available, (v) expression of the cDNAs obtained from the Clontech library of document D37 per se did not address the validity of the cDNA clones obtained, (vi) in the absence of authentic DNA sequence data, genomic gene cloning and characterisation would have imposed an undue burden on the skilled person, and (vii) the prior art provided no guidance regarding which, if any, of these routes to pursue, the skilled person would have encountered technical difficulties that he or she would have been unable to surmount by routine experimentation.

XIII. The respondent did not make any submissions or requests in the appeal proceedings.

XIV. The appellant requested that the decision under appeal be set aside and that the patent be maintained on the basis of either the main request filed on 28 February 2001 for all designated Contracting States except ES.
and corresponding claims for the Contracting State ES filed during the oral proceedings or auxiliary request A or B filed on 31 October 2003.

**Reasons for the Decision**

*Main request for all designated Contacting States except ES*

**Article 123(2) and Rule 88 EPC**

**Amendments in Table 1**

1. Table 1 in the application as filed (see page 6) provides a list of the differences distinguishing the amino acid sequence of the invention from the amino acid sequence of Figure 1 of document D2. Table 1 in the patent (see page 1) differs therefrom in that three differences are no longer mentioned, namely the three amino acid substitutions occurring at positions 155, 156 and 321 (according to the numbering of the sequence of Figure 2 of the application/patent).

2. The codons which correspond to the amino acid residues at positions 155, 156 and 321 in Figure 2 of the application as filed are TTC, TTG and AAG which code for Phe, Leu and Lys, as correctly indicated in the Figure. Looking in Figure 1 of document D2 for alignments consisting of those three amino acid residues together with their flanking amino acid residues as represented in Figure 2 of the application as filed, the skilled person would find that the same three amino acid residues are also present in Figure 1.
of document D2 at positions 135, 136 and 300, respectively.

3. The skilled person would conclude that Table 1 in the application as filed is erroneous and would regard removal therefrom of any references to those three "differences" as the only possible correction, and as being obvious within the meaning of Rule 88 EPC.

Amendment in Figure 2

4. In Figure 2 of the application as filed the amino acid residue at position 296 is Gln, whereas in Figure 2 of the patent it has been replaced by Glu.

5. Because the codon corresponding to position 296 is GAA which codes for Glu the skilled person would realize immediately that in Figure 2 as filed either the codon (GAA) or the indicated amino acid (Gln) is erroneous.

6. As the inventors have first experimentally identified the cDNA molecule and only in a second step, using a reasoned approach based on a reading of the genetic code, deduced therefrom the sequence of the encoded protein, the person skilled in the art would conclude that not the codon but the amino acid residue is erroneous and would regard replacement at position 296 in the Figure of Gln by Glu, as the only possible correction, and as being obvious within the meaning of Rule 88 EPC.
The feature "derived from" in claim 1

7. Claim 1 is directed to a cDNA coding for an iron-binding protein derived from a human lactoferrin, which cDNA comprises a sequence coding for the amino acid sequence 74 to 275 of Figure 2. As the wording "derived from" is not explicitly used in the application as filed, it has to be assessed whether an implicit support can be found for this feature in the application as filed.

8. Page 9, lines 12 to 18 in the application as filed points to a polypeptide shorter than the entire human lactoferrin, a preferred polypeptide consisting of the region delimited by amino acids 74 and 275 which contains an iron binding domain. Claim 8 as filed which reads: "8. A cDNA sequence comprising a portion of the cDNA of Fig. 2 coding for human lactoferrin protein including at least one of the iron binding domains with an Fe binding site." corresponds with that passage of the description.

9. Both that passage and that claim provide a clear indication that not only the entire protein but also any derivatives thereof, provided that they include the region delimited by amino acids 74 and 275, as well as the corresponding cDNAs are part of the invention as described in the application as filed.

10. Therefore, a cDNA coding for an iron-binding protein derived from a human lactoferrin is disclosed in the application as filed.
11. The main request meets the requirements of Article 123(2) EPC.

Article 87 EPC (entitlement to the priority date)

12. As the description and drawings of the priority document and the description and drawings of the application as filed have the same content and as, furthermore, claim 1 as filed has the same content as claim 1 of the priority document and claims 3 and 5 as filed are identical with claims 2 and 3 of the priority document, the skilled person could derive the subject-matter of claims 1 to 10 of the main request directly and unambiguously from the previous application as a whole. Therefore, in accordance with decision G 2/98 (OJ EPO 2001, 413), the requirement for claiming priority of the same invention referred to in Article 87(1) EPC is met. Thus, the main request is entitled to the priority date of 5 May 1989.

Article 83 EPC (sufficiency of disclosure)

13. Figure 2 of the patent provides both the complete sequence of the cDNA as retrieved by the inventors and the complete amino acid sequence encoded thereby. Figure 1 clearly indicates the different portions of the nucleotide sequence respectively encoding the peptide signal and the mature protein as well as length of the corresponding amino acid sequences. Therefore, a clear and complete disclosure of the claimed subject-matter is provided by the patent specification. Thus, the main request meets the requirements of Article 83 EPC.
Article 54 EPC (Novelty)

14. In the decision under appeal it was considered that the subject-matter of claims 7 to 9 was not new over document D3. In the course of the opposition it was also argued that the cDNA and the corresponding amino acid sequence of the invention (all claims concerned) had been disclosed at a conference held before the priority date.

Document D3

15. In document D3 the results of an x-ray analysis of the structure of human lactoferrin at 3.2-Å resolution are reported. The results allowed a description of the polypeptide chain folding, and the nature and location of the iron binding sites. It was taken for granted by the authors that the tested lactoferrin which had been isolated from human milk had the amino acid sequence disclosed in Figure 1 of document D2 which significantly differs from the amino acid sequence of Figure 2 of the patent (see Table 1 on page 5 of the patent).

16. The mere statement in the bottom of the left-hand column of page 1770 of document D3 which reads: "In the final tracing there are two significant breaks in the chain, both in the C-terminal lobe (residues 388-403 and 429-433 have no density). The N-terminal residues 1-5 are also not visible." reflects doubts about the correctness of certain portions of the amino acid sequence of document D2. Nevertheless, nowhere in document D3 is there in this respect any correction suggested and a fortiori any corrected sequence.
described. Therefore, document D3 cannot deprive the subject-matter of any of claims 7 to 9 of novelty.

Oral disclosure (San Antonio Meeting)

17. As evidenced in document D31, a declaration of Dr Rado, and in document D32, a declaration of Dr Wei, a lecture was given by Dr. Rado at a meeting held at San Antonio, Texas, USA, in December 3-6, 1988 ("San Antonio Meeting"). Dr Rado spoke in place of Dr Wei who originally was intending to give the presentation (see point 7 of document D31).

18. In document D31 (see points 8 and 12 thereof), Dr Rado declared inter alia: "During the presentation, I described the strategy for cloning pHL-44, and thereby obtaining the full coding sequence of human lactoferrin cDNA and the corresponding amino acid sequence. I specifically recall presenting a slide showing the full-length cDNA and amino acid sequence of lactoferrin that we had determined. The sequences I presented were the same sequences as later submitted to Genbank (Exhibit E). These are also the same sequences (including the minor corrections discussed in (4)) shown in Exhibit C. I also presented a slide showing the restriction map of one of the genomic human lactoferrin clones described above (Exhibit F).", and "During the presentation, I had with me numerous paper copies of the slide showing the cDNA and amino acid sequences of human lactoferrin that we had determined. As I was giving the presentation, I told the audience that I would make available paper copies after the presentation, and I did distribute several copies to audience members.".
19. No reproductions of the slides presented by Dr Rado, in particular of the one showing full-length cDNA and amino acid sequence of human lactoferrin are annexed to either of documents D31 and D32. Nor are copies provided of the papers handed out to the members of the public. The only detailed information relating to cDNA and amino acid sequences is contained in two exhibits which are **Exhibit B** to each of documents D31 and D32 and **Exhibit C** to document D31:

19.1 **Exhibit B** represents the first print out of the cDNA sequence obtained from two cDNA clones, retrieved by Dr Rado and Dr Wei and designated pHL-41 and pHL-44, containing overlapping sequences spanning the full coding sequence of human lactoferrin. Apart from the fact that that cDNA sequence differs in many places from the cDNA sequence as represented in Figure 2 of the patent, nothing in either of documents D31 and D32 indicates that this sequence was that depicted on the slide allegedly shown by Dr Rado.

19.2 **Exhibit C** shows a corrected version of the cDNA sequence of Exhibit B as well as the corresponding amino acid sequence. Apart from the fact that those sequences differ in many places from the sequences as represented in Figure 2 of the patent, Dr Rado's declaration does not state explicitly that this was the actual slide shown at the meeting. It merely states that these sequences were the same submitted to GenBank at a later date (after the priority date) which in turn were the same as those presented at the meeting. This is not considered to be an "up to the hilt" proof of
what was actually shown or distributed by Dr Rado at the meeting.

20. Moreover, the mere contentions by Dr Rado and Dr Wei in their declarations that the disclosure occurred cannot be accepted because both of them, Dr Rado as being the lecturer and Dr Wei as being the person who prepared the speech, are not qualified to provide evidence safely and satisfactorily establishing the information content made publicly available by the lecture and what an ordinary member of the audience at the lecture would have understood (see decision T 1212/97, supra, points 4 and 5 of the reasons).

21. Therefore, the board concludes that no evidence has been provided showing that the claimed subject-matter was disclosed at the San Antonio Meeting.

22. Therefore, the main request meets the requirements of Article 54 EPC.

Article 56 EPC (inventive step)

23. Having regard to the state of the art as illustrated hereinafter (see point 24, infra), the technical problem faced by the inventors of the patent at issue was the provision of a complete cDNA encoding the genuine human lactoferrin to be used - as stated in the patent specification - for preparing synthetic human lactoferrin in a eucaryotic expression system. The solution thereto is the particular cDNA of claims 1 to 6 which is represented in Figure 2 of the patent and the synthetic proteins produced therewith in a eucaryotic expression system.
24. The relevant state of the art the skilled person would have been aware of are documents D1, D2, D3 (see points 15 and 16, supra) and D37, i.e. the four documents which have been taken into consideration in the decision under appeal, as well as document D8. The skilled person would have made the following analysis of those documents and, thus, of the state of the art in the field of human lactoferrin:

24.1 **Document D1**, to which both Dr Rado and Dr Wei contributed, describes isolation of clone pHL-41. That cDNA clone encodes an amino acid sequence (see Figure 1) which corresponds to the major part (residues 428 to 703) of the C-terminus of the amino acid sequence reported in the earlier document D2 (to which reference is made) and differs therefrom in three residues. This unique partial cDNA was obtained by screening a cDNA library which had been prepared from a human myeloid leukemia cell line.

24.2 **Document D2** deals with the determination of the amino acid sequence of human lactoferrin. The nature of the starting material from which the protein has been purified is not indicated. The complete sequence of the mature protein is given in Figure 1 (see page 661). Prior to the present invention, this was considered to be the very sequence of human lactoferrin.

24.3 **Document D3**, while reporting results of an x-ray analysis of the structure of human lactoferrin, indirectly expresses doubts as to the correctness of the first five amino acids and amino acids comprised between position 388 and 433 of the sequence of
document D2, i.e. partly in the N-terminal portion of the polypeptide encoded by the cDNA clone pHL41 of document D1.

24.4 **Document D8**, to which both Dr Rado and Dr Wei contributed, reports on the isolation and characterisation of two cDNA clones, referred to as pHL41 and pHL44, which comprise the full length coding sequence of the gene encoding human lactoferrin. While clone pHL41 is characterised by reference to document D1, clone pHL44 is said to have been isolated from a library specifically primed with a 19-mer oligonucleotide containing a sequence complementary to a nucleotide stretch of pHL41. This library was screened with a 90 bp PstI-EcoRV fragment representing the most upstream sequence of pHL41. Both strands of pHL44 were sequenced and it was found that it contained the entire coding sequence of human lactoferrin. The sequences of the two clones are not described.

24.5 **Document D37** is a "product analysis certificate" from Clontech Laboratories Inc. which provides brief information about a human breast cDNA library, designated with the catalog number "HL 1037a". The library was prepared from a breast tissue of a female showing lactational competence. No information concerning the mRNA present in the cells from which the cDNA library was prepared was made available. Document D37 is undated. But based on document D34 which consists of the cover page of the Clontech catalog, headed "1988 CLONTECH Products and Protocols Catalog" and page 1.9 thereof which refers *inter alia* to the library with catalog number "HL 1037a", it can be
assumed that the information contained in document D37 had been made available to the public in 1988.

25. The skilled person would have regarded document D8, which represents a further development of document D1, as the most relevant document. He or she would have attempted to construct a cDNA library as indicated in document D1. Then, using the probe and the 90 bp PstI-EcoRV fragment referred to in document D8, both being easily available from the pHL41 sequence of Figure 1 of document D1, he or she would have screened the cDNA library in an attempt to identify the clone pHL44. In this respect, it may be observed that, being a cautious person, he or she certainly would not have embarked on the screening of the cDNA library of document D37, because, on the one hand, he or she would have had no reason to doubt whether a relevant cDNA library could be prepared as indicated in document D1 and, on the other hand, he or she would have had absolutely no information concerning the cDNAs of the library of document D37, in particular as to the polypeptides encoded thereby.

26. However, as authoritatively shown by both the declaration of Dr Wei (document D32) and the declaration of Dr Stowell (document D49), the skilled person would have inevitably failed in his or her attempt. Indeed, not only was the information delivered by document D8 that the cDNA clone pHL44 contained the entire sequence coding sequence of human lactoferrin false (see point 6 of document D31), but also even if he or she had succeeded in isolating the actual clone pHL44 (as characterised in document D31) he or she would have obtained an imperfect cDNA incapable of
expressing the encoded protein (see points 4 and 5 of the declaration of Dr Stowell (document D49)).

27. Preparation of a cDNA as defined in any of claims 1 to 6 would have required different routes of experimentation which were not described in the state of the art at the priority date, such as those followed by the inventors or those reported in the post-published document D42 (to which Dr Rado contributed) which rely on the synthesis of a cDNA from bone-marrow RNA and the cloning of that cDNA in baby-hamster kidney cells.

28. Therefore, in the board's judgment, it has to be concluded that, in spite of the prima facie wealth of information in the state of the art about human lactoferrin, it was not obvious for the skilled person to arrive at the subject-matter of claims 1 to 6.

29. As the structure of the proteins according to claims 7 to 9 could be established only by inference from the cDNA according to claims 1 to 6, and as claim 10 is directed to a method of producing such a protein by expressing such a cDNA, these other aspects of the claimed invention also involve an inventive step. Thus, the main request as a whole meets the requirements of Article 56 EPC.
Claims for the Contracting State ES

30. The same conclusions apply to the claims for ES.

Description

31. The board sees no need to amend the description to take account of the amendments in the claims of the main request for all designated Contracting States except ES as these were simply the deletion of granted claims 11 and 12 which contained the term "C-lobe" for which it was found by the opposition division that there was no support in the application as filed (see point 2 of the opposition division's communication of 11 September 1997).
Order

For these reasons it is decided that:

1. The decision under appeal is set aside.

2. The case is remitted to the first instance with the order to maintain the patent on the basis of the claims of the main request filed on 28 February 2001 for all designated Contracting States except ES and corresponding claims for the Contracting State ES filed during the oral proceedings, and the description and drawings as granted.

The Registrar:  The Chairman:

A. Wolinski     L. Galligani