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D E C I S I O N
of 4 December 2002

Case Number: T 1099/99 - 3.3.8

Application Number: 86306624.7

Publication Number: 0215594

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Title of invention:
Heterologous polypeptide expressed in filamentous fungi, processes for their preparation, and vectors for their preparation

Patentee:
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Opponents:
(01) F. HOFFMANN-LA ROCHE & CO. Aktiengesellschaft
(02) Agennix Incorporated
(04) Unilever N.V.
(05) WACKER-CHEMIE GmbH
(06) Primalco Ltd.

Headword:
Filamentous fungi/GENENCOR

Relevant legal provisions:
EPC Art. 54, 56, 87, 88, 89

Keyword:
"Main request - novelty (no)"
"First auxiliary request - novelty (yes)"
"Inventive step (no)"

Decisions cited:

G 0009/92, T 0206/83, T 0060/89, T 0158/91, T 0782/91,
T 0207/94, T 0351/98, T 0338/00

Catchword:
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Case Number: T 1099/99 - 3.3.8

D E C I S I O N
of the Technical Board of Appeal 3.3.8
of 4 December 2002

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Decision under appeal: **Interlocutory decision of the Opposition Division of the European Patent Office posted 14 October 1999 concerning maintenance of European patent No. 0 215 594 in amended form.**

Composition of the Board:

Chairman: L. Galligani
Members: P. Julia
S. C. Perryman

Summary of Facts and Submissions

I. The appeal lies from the interlocutory decision of the opposition division issued on 14 October 1999 concerning the maintenance of the European patent No. 0 215 594 in amended form. The patent had been granted on the basis of claims 1 to 29, wherein independent claim 1 read as follows:

"1. A process for making a heterologous polypeptide comprising : transforming a filamentous fungus from the subdivision Eumycotina with a vector containing DNA sequences capable of expressing a heterologous polypeptide and of causing secretion of the heterologous polypeptide from the filamentous fungus, said filamentous fungus being selected from members of the subdivision Eumycotina that are capable of being propagated in filamentous form, and expressing and secreting said heterologous polypeptide."

Dependent claim 2 defined the DNA sequences comprised in the vector (encoding the heterologous polypeptide, encoding a signal sequence and a promoter sequence), whereas dependent claims 3 to 29 further defined the signal sequences, promoter sequences, further elements in the vector, such as functional transcription termination and polyadenylation sequences, DNA sequences encoding a selection characteristic expressible in filamentous fungus, DNA sequences capable of increasing the transformation efficiency, the secreted heterologous polypeptide and the filamentous fungi.

II. The granted patent had been opposed by six opponents (opponents 01 to 06) under Article 100(a), (b) and (c)

EPC, of which one later withdrew its opposition.

The opposition division, while not allowing the main request (claims as granted) for lack of novelty, decided that the subject-matter of the fifth auxiliary request then on file fulfilled the requirements of the EPC.

Independent claim 1 of this request read as follows:

"1. A process for making a mammalian polypeptide comprising: transforming a filamentous fungus from the subdivision Eumycotina, with the exclusion of Saccharomyces cerevisiae, that are capable of being propagated in filamentous form with a vector which comprises a DNA sequence encoding said polypeptide, a DNA sequence encoding a signal sequence and a promoter sequence operably linked to said DNA encoding the signal sequence, said promoter sequence being functionally recognized by said filamentous fungus, whereby said DNA sequences are capable of expressing said polypeptide and of causing secretion of the polypeptide from the filamentous fungus, and expressing and secreting said polypeptide; wherein said signal sequence is native to the mammalian polypeptide or comprises the signal sequence of bovine preprochymosin or Mucor miehei preprocarboxy protease."

Independent claim 2 was essentially the same as claim 1 but directed to a process for making a heterologous polypeptide wherein the signal sequence comprised the signal sequence of bovine preprochymosin or Mucor miehei preprocarboxy protease. Dependent claims 3 to 18 defined particular embodiments of the process of either claim 1 or 2.

III. Both the patentee and opponent 02 lodged an appeal against the interlocutory decision of the opposition division. Opponent 02 withdrew its appeal and its opposition on 7 April 2000, whereas opponent 06 withdrew its opposition on 14 February 2001.

IV. On 28 August 2002, the board issued a communication pursuant to Article 11(2) of the rules of procedure of the boards of appeal indicating with reference to decision G 9/92 (OJ EPO 1994, 875) that neither the board of appeal nor the non-appealing opponents might challenge the maintenance of the patent as amended in accordance with the interlocutory decision.

V. In reply to the board's communication, respondent II (opponent 04) and respondent III (opponent 05) informed the board of their intention not to attend the oral proceedings. The appellant (patentee) filed several auxiliary requests (auxiliary request A to G and a "Penultimate Auxiliary Claim Request") and written submissions with additional prior art. No submissions in writing were made by any of the respondents.

VI. Oral proceedings were held on 4 December 2002. During oral proceedings the appellant withdrew all its previous requests and filed a new main request with claims 1 to 28 and a first auxiliary request with claims 1 to 3. Independent claim 1 of the new main request read as follows:

"1. A process for making a heterologous polypeptide comprising : transforming a filamentous fungus from the subdivision Eumycotina with a vector containing DNA sequences capable of expressing a heterologous polypeptide and of causing secretion of the

heterologous polypeptide from the filamentous fungus, the vector comprising a DNA sequence encoding said heterologous polypeptide, a DNA sequence encoding a signal sequence and a promoter sequence operably linked to said DNA encoding the signal sequence, said promoter sequence being functionally recognized by said filamentous fungus, said filamentous fungus being selected from members of the subdivision Eumycotina that are capable of being propagated in filamentous form but excluding yeasts, and expressing and secreting said heterologous polypeptide."

Dependent claims 2 to 28 corresponded to the subject matter of claims 3 to 29 of the granted claims, with the deletion of Trichoderma reesei in the last claim.

Independent claim 1 of the first auxiliary request read as follows:

"1. A process for making a mammalian polypeptide comprising : transforming a filamentous fungus from the subdivision Eumycotina with a vector containing DNA sequences capable of expressing a heterologous polypeptide and of causing secretion of the heterologous polypeptide from the filamentous fungus, the vector comprising a DNA sequence encoding said mammalian polypeptide, a DNA sequence encoding a signal sequence and a promoter sequence operably linked to said DNA encoding the signal sequence, said promoter sequence being functionally recognized by said filamentous fungus, said filamentous fungus being selected from members of the subdivision Eumycotina that are capable of being propagated in filamentous form but excluding yeasts, and expressing and secreting said heterologous polypeptide; wherein said signal

sequence is native to the mammalian polypeptide or is derived from bovine preprochymosin, Mucor miehei preprocarboxy protease or a Trichoderma reesei cellulase."

Independent claim 2 was directed to a process for making a heterologous polypeptide from Humicola or Mucor species with a signal sequence native to the heterologous polypeptide and using an Aspergillus filamentous fungus as a host. Independent claim 3 was essentially as claim 1 but directed to a process for making a heterologous polypeptide with a signal sequence derived from bovine preprochymosin, Mucor miehei preprocarboxy protease or a Trichoderma reesei cellulase.

VII. The following documents are cited in this decision:

D1: US application 664 230 (priority document of D2);

D2: EP 0 191 221 (A1);

D4: EP 0 244 234 (A2);

D5: GB application 86 10600 (priority document of D4);

D6: EP 0 099 226 (A1);

D7: M.A. Innis et al., Science, 5 April 1985, Vol. 228, 21 to 26;

D10: A.-M. Bech and B. Foltmann, Neth. Milk Dairy J., 1981, Vol. 35, 275 to 280;

D12: D.J. Ballance et al., Biochem. Biophys. Res.

Commun., 1983, Vol. 112 (1), 284 to 289;

D13: J.M. Kelly and M.J. Hynes, The EMBO J., 1985, Vol. 4 (2), 475 to 479;

D15: M.M. Yelton et al., Proc. Natl. Acad. Sci. USA, 1984, Vol. 81, 1470 to 1474;

D16: D.J. Ballance, Abstract presented at the EMBO-Workshop April 17 to 19, 1984, Rhenen, The Netherlands;

D23: D.J. Ballace and G. Turner, Gene, 1985, Vol. 36, 321 to 331;

D34: WO 86/03774 (A1);

D35: DK application 6019/84 (priority document of D34);

D47: EP 0 137 280 (A1);

D59: T. Taniguchi et al., Proc. Natl. Acad. Sci. USA, 1980, Vol. 77, 5230 to 5233;

D60: J. Mellor et al., Gene, 1983, Vol. 24, 1 to 14.

VIII. The arguments of respondent I (opponent 01) for the main request can be summarized as follows: (i) claims concerned with Trichoderma (claims 26 and 27) are not entitled to the first priority date of the contested patent because the first priority document is not enabling for Trichoderma. Document D4 (with document D5 as a valid priority), which discloses the methods of the contested patent in Trichoderma, anticipates the subject matter of those claims. (ii) Having regard to

documents D2 (with document D1 as a valid priority), D6 and D34 (with document D35 as a valid priority), the main request lacks novelty.

The arguments for the first auxiliary request can be summarized as follows: (i) the introduction of the signal sequence of the Trichoderma reesei cellulase into the claims is not occasioned by any ground for opposition and thus, it cannot be allowed under Rule 57(a) EPC. (ii) The claimed subject matter lacks inventive step, in particular in view of document D47 in combination with document D6 for claim 3 and document D10 in combination with document D6 for claim 2.

IX. The argumentation of the appellant for the main request is essentially as follows: (i) the first priority document is enabling for Trichoderma. Document D4 is not relevant for claims 26 and 27 as they are entitled to said priority. (ii) Document D2 discloses a complementation system with expression and secretion of a homologous polypeptide. There is no production of a heterologous polypeptide in the sense defined in the patent-in-suit. The references to the secretion of heterologous polypeptides in document D2, like the ones in documents D6 and D34, are mere speculations without any reliable technical basis. Neither document D2 nor any other document of the cited prior art enables the skilled person to carry out these suggestions in a straightforward manner. In view of the difficulties and uncertainties shown in the prior art, such as the failures reported in documents D59 and D60, the skilled person would not seriously contemplate following these suggestions. Thus, none of the said documents affects novelty.

The arguments in support of the first auxiliary request are essentially as follows: (i) the introduction of the signal sequence of the Trichoderma reesei cellulase into the claims overcomes the novelty objection raised for the main request and thus, the requirements of Rule 57(a) EPC are fulfilled. (ii) Document D47 only discloses the expression and secretion of the Trichoderma reesei cellulase using yeast as a host. There is no suggestion, let alone any motivation, for selecting a filamentous fungus as a host. Making such a selection entails an unacceptable degree of hindsight. Moreover, even if it were obvious for the skilled person to make such a selection, he or she would have no reasonable expectation of success in view of the problems and the uncertainties shown in the cited prior art (as outlined for the main request).

X. The appellant requested that the decision under appeal be set aside and the patent maintained on the basis of the main request or of the first auxiliary request, both submitted at the oral proceedings on 4 December 2002.

The respondents requested that the appeal be dismissed.

Reasons for the Decision

Main request
Articles 123(2), (3) EPC and 84 EPC

1. Claim 1 of this request is a combination of granted claims 1 and 2 with the exclusion of yeast. This exclusion has a basis in the description as originally filed (cf page 4, lines 40 to 41) and it is a

restriction of the granted claims by excluding those yeasts from the subdivision Eumycotina which under certain conditions are capable of being propagated in filamentous form. The objections originally raised under Article 123(2) EPC against the granted claims only concerned dependent claims 23 to 25. The findings of the decision under appeal have not been disputed by the respondents. The board sees no reason to question these findings. Thus, the main request meets the requirements of Articles 123(2), (3) EPC and 84 EPC.

Articles 87 to 89 EPC (Entitlement to priority)

2. Claims 26 and 27 concern inter alia the particular embodiment of the process of claim 1 in which Trichoderma is used as a host. Formal support for the use as hosts of several filamentous fungi, including Trichoderma, is found in the first priority document (cf page 9, lines 29 to 31, claim 27). However, respondent I has argued that at the priority date no method was available for transforming Trichoderma and that example 7 of the patent-in-suit shows that the method used for transforming Aspergillus, which is the only one disclosed in the first priority document, requires several modifications in order to be applied to Trichoderma. For this reason, in its view, this aspect of the claims is not entitled to the first priority date (29 August 1985).

No objections have been raised in respect of other filamentous fungi cited in the patent-in-suit or in the first priority document.

Thus, it has to be established whether the information given in the first priority document as a whole, and

possibly supplemented by the common general knowledge, would enable the skilled person to carry out the claimed process using Trichoderma filamentous fungi as a host (cf eg T 351/98 of 15 January 2002, see point 39 of the reasons).

2.1 At this stage it is highly relevant to consider the disclosure of the first priority document as a whole and in particular the following general teachings:

(i) the culture conditions used for Aspergillus nidulans transformants are disclosed on page 15 of the first priority document (minimal agar media with sodium nitrate). Example 1 (page 19) refers to normal culture conditions for A. niger (potato dextrose broth, 30°C) and example 6 (page 35) to the conditions for Mucor miehei (YMB medium with yeast extracts). Thus, the skilled person is made aware of something which is already obvious in the art, namely that each and every fungus has its optimal conditions for culture and that for one and the same fungus different culture conditions can be used. The explicit references to the use of other filamentous fungi (page 9) are addressed to the person skilled in the art, who in this case would have (or would be in a position to easily acquire) general knowledge on Trichoderma, including suitable and optimal conditions for culture.

(ii) the method for transformation of Aspergillus disclosed on pages 13 to 14 of the priority document is said to be based on document D12 with several modifications. In particular, the hydrolytic enzyme mixture Novozyme 234 used to

digest the mycelia cell walls (which is known to have a variable quality depending on the commercial lot used) is first partially purified so as to provide a greater amount of protoplasts and a higher frequency of stable transformants. The first priority document further discloses several vectors (pGRG1-pGRG4) with improved transformation frequencies (by presence of the ANS-I sequence). Thus, the skilled person is made aware of low transformation frequencies and instructions and means are provided in the priority document for overcoming this problem and improving the transformation frequencies.

2.2 In view of the fact that the process claimed in the main request only requires the transformation of filamentous fungi but without requiring any specific yield or transformation frequency, the board is convinced that the information provided in the first priority document as a whole (with common general knowledge) would enable the skilled person to achieve Trichoderma protoplasts, their successful transformation as well as suitable culture conditions for Trichoderma. The modifications referred in example 7 of the patent-in-suit are seen as normal modifications that the skilled person could easily achieve (or similarly suitable ones) with the information disclosed in the first priority document and the common general knowledge. Thus, the first priority document is considered to be enabling for Trichoderma.

Success with other fungi referred to in the contested patent has not been challenged by the respondents and, in view of the arguments given above, neither does the

board see any reason to doubt that these can be used as claimed.

2.3 Thus, the main request is entitled to the first priority date (29 August 1985).

Article 54(3)(4) EPC (Novelty)

3. Document D2 is a document cited against novelty under Article 54(3) EPC with document D1 as a valid priority (24 October 1984). Document D2 refers to filamentous (ascomycetes) fungi as "natural" secreters (page 4, lines 16 to 22) and provides plasmid and cosmid vectors as intermediate products which allow to obtain fungal signal sequences for the secretion of proteins encoded by selected foreign DNA sequences linked in suitable reading frame (page 4, lines 11 to 16). References are found to "control sequences" which control the expression of these operably linked coding sequences (page 6, line 26 to page 7, line 3). The general teaching of this document comprises the use of these vectors for the efficient production and secretion of heterologous polypeptides by mediation of suitable fungal signal sequences (page 14, lines 9 to 24). No technical differences can be seen between this teaching and the process claimed in the main request.

According to the established case law of the boards of appeal, a document is only considered to be relevant for novelty purposes if its disclosure is an enabling disclosure (cf eg T 206/83, OJ EPO 1987, 5). Thus, the key question is whether document D2 provides sufficient information for the skilled person to carry out the above referred general teaching in a straightforward manner with common general knowledge, ie whether

document D2 is an "enabling disclosure" or not.

3.1 Secretion of a polypeptide

Document D2 exemplifies the construction of the plasmid pHY201 and the cosmid pKBY2 (example C, page 16 to page 23 and example D, page 23 to page 29, respectively). Plasmid pHY201 is derived from the cloning vector pBR329 with a suitable Aspergillus nidulans selection-marker (trpC gene). This plasmid successfully transforms A. nidulans protoplasts and it is integrated in Aspergillus transformants. Cosmid pKBY2 is constructed in a similar manner with the additional presence of cos sites. This pKBY2 cosmid is further used as a cloning vector for an Aspergillus genomic library resulting in the isolation of a 35-40 kb fragment which comprises the yA2 gene encoding the extracellular enzyme p-diphenol oxidase (laccase). The presence of the yA2 gene is confirmed by detecting the conidial yA2 laccase in protein extracts and by the transformation of a yellow spored ($yA2^-$) A. nidulans strain and reversion to a green conidia $yA2^+$ phenotype. The reversion of the yellow conidia phenotype indicates that the extracellular (secreted) laccase performs its normal biological function.

3.2 Secretion of a heterologous polypeptide

The appellant has argued that document D2 does not disclose the secretion of a heterologous polypeptide in the sense of the patent-in-suit. The board, however, cannot share this view since the definition of "heterologous polypeptide" found in the contested patent does not exclude a homologous complementation. It is true that in referring to the prior art the

specification distinguishes between "homologous fungal expression" (involving complementation) and "heterologous fungal expression" (page 3, lines 37 to 54). However, on page 5, lines 28 to 29 under the heading "Definitions", the "heterologous polypeptides" are defined as polypeptides which are not normally expressed and secreted by the filamentous fungus used to express these polypeptides. Further on lines 33 to 34 reference is made to polypeptides derived from fungal sources other than the expression host. Thus, what is excluded by the definition of "heterologous polypeptide" is very narrow and restricted to the very specific individual host strain used, even if the examples given are all at the level of species. A polypeptide which is not produced by a specific strain must therefore be seen as a "heterologous polypeptide" to this specific strain. This interpretation is further confirmed on lines 45 to 47, wherein the "heterologous polypeptides" are said to include "naturally occurring allelic variations that may exist or occur in the sequence of polypeptides derived from the above ... fungal sources ...". As far as the "above" referred "heterologous polypeptides" are not clearly defined, the expression and secretion of a specific allelic variant using as a host a filamentous fungus strain which (normally) does not produce such an allelic variant is seen as embraced by the claimed process too. In view of this interpretation, document D2, which discloses the complementation of a yA2$^-$ strain (not normally expressing and secreting the yA2 laccase) by the (heterologous) yA2 gene, is considered to anticipate the subject matter of the main request.

3.3 The general teaching of document D2 is enabling

Even if, for the sake of argumentation, the example given in document D2 were to be considered as a disclosure of a "homologous polypeptide" in the sense given by the appellant, the teachings of document D2 clearly refer to the use of the disclosed vectors for the efficient expression and secretion of "heterologous polypeptides" such as, for example, insulin or other hormones, lymphokines, growth factors, or other enzymic or structural proteins (page 15, lines 32 to 34). Document D2 provides straightforward and sufficient instructions for the skilled person to carry out this general teaching without undue burden, only routine experimentation and trials being involved and the desired results being directly verifiable (expression and secretion). No technical evidence has been put forward to contradict this. To accept the appellant's argument that document D2 is not enabling as far as the general teaching is concerned would be to apply a different standard to the disclosure of this prior art than to the patent in suit (not limited to any filamentous fungi, transforming vector, signal sequence, etc...) which would be contrary to the established case law (cf T 158/91 of 30 July 1991 and T 782/91 of 12 December 1994).

3.4 In the board's judgment, the gist of the invention as disclosed in the patent-in-suit is already found in document D2 and, as stated above, this document provides methods and means enabling the skilled person to perform it. Contrary to the appellant's argumentation, the general teaching of document D2 (which is essentially the same as the one of the patent in suit) as well as the general and specific products for performing this teaching are considered to "make available" the subject matter of the contested patent.

3.5 Thus, claim 1 of the main request is considered to lack novelty over the disclosure of document D2 (Article 54(3), (4) EPC) and consequently, the main request, which comprises it, is not found to satisfy the requirements of the EPC.

First auxiliary request
Articles 123(2), (3) EPC and 84 EPC. Rule 57(a) EPC

4. The three independent claims of this first auxiliary request are particular combinations of the granted claims with a formal basis in the claims and the description as originally filed (see point 1 above). The wording relative to the signal sequence of the Trichoderma reesei cellulase is found on page 5, lines 18 to 20 of the application as originally filed. The claimed subject matter essentially amounts to a limitation to specific embodiments of the granted claims. Moreover, the limitation to specific signal sequences overcomes the objection of lack of novelty over document D2. Thus, the first auxiliary request is considered to fulfil the requirements of Articles 123(2), (3) EPC and 84 EPC as well as those of Rule 57(a) EPC.

Article 54 EPC (Novelty)

5. No document on file discloses a process for making a mammalian or a heterologous polypeptide using the specific signal sequences cited in claims 1 or 3 and a filamentous fungus as a host. The production of an Humicola or Mucor polypeptide with its native signal sequence using the filamentous fungus Aspergillus as a host (claim 2) is not anticipated by the cited prior art. Thus, novelty is acknowledged (Article 54 EPC).

Article 56 EPC (Inventive step)

6. This request contains three independent process claims which essentially comprise similar technical features. Independent claim 3, being a process for making a general heterologous polypeptide, is broader than independent claim 1 which is directed to a similar process for making a mammalian polypeptide. Independent claim 2 is directed to a similar process for making a heterologous polypeptide from Humicola or Mucor species.

7. The closest prior art to the subject matter of claim 3 of this request is considered to be document D47. This document is concerned with the production of recombinant cellobiohydrolases (CBH) or cellulase enzymes from fungal sources, more particularly the CBH enzymes (CBHI and CBHII) from Trichoderma reesei. Document D47 explicitly refers to signal sequences (page 10, line 29 to page 11, line 3) and to the use of several hosts, including general eucaryotic microbes, such as yeast (page 14, lines 3 to 4). The document exemplifies the expression and secretion of the Trichoderma reesei CBHI cellulase in Saccharomyces cerevisiae using a signal sequence derived from the CBHI cellulase gene (example D.10, pages 59 to 61).

8. Starting from this closest prior art, the objective technical problem underlying the contested patent must be seen in the provision of alternative hosts for expressing and secreting these cellulase enzymes. Claim 3, which in its broad outline covers a method for expressing in a filamentous fungus Trichoderma reesei cellulase with its own signal, provides a solution to said problem.

9. Document D47 not only refers to the production of these cellulase enzymes in other hosts but it further contemplates the use of the disclosed CBH genes for modifying or altering (by way of inactivation or enhancement) the ratio or the amount of cellulases produced by a general organism. In this context, reference is made to fungal organisms and to document D15, which is cited for demonstrating the feasibility of this approach in filamentous fungal organisms (page 5, lines 9 to 24). Document D15 discloses the transformation and complementation of an Aspergillus nidulans trpC$^-$ strain with a trpC gene from A. nidulans and it further refers to other transformation results in Aspergillus (cf document D12) and in Neurospora crassa (by the inventors of document D6) (page 1474, left-column). In view of this pointer and knowing the wide industrial use of filamentous fungi (production of enzymes in food industry), it would have been obvious for the skilled person to follow this indicated path and select the filamentous fungi as an alternative host to the exemplified yeast. The relevant question is thus whether the skilled person, based on a scientific evaluation of the facts at hand, would have had a reasonable expectation of success (cf T 60/89 OJ EPO 1992, 268).

10. The appellant has indicated several factors that would put in jeopardy any reasonable expectation of success in particular in respect of the possibility of achieving secretion from the host. However, as stated for eg in T 207/94 (OJ EPO 1999, 273, see point 34 of the reasons), an assumption or hypothesis about a possible obstacle to the successful realisation of a project, in order to be considered, must always be

based upon facts. In the present case and for the reasons given hereinafter, the board is of the opinion that none of these alleged factors would have lowered the expectations of the skilled person.

It is true, as rightly emphasized by the appellant, that even if filamentous fungi are known to be natural (good) secreters, the actual yield of secretion can be influenced by many factors such as (i) the medium used (presence of proteases, amount of nutrients and growth factors, etc...), (ii) the nature of the heterologous polypeptide (presence of a feedback with the medium, ie whether or not the polypeptide is important for degrading the nutrients present in the medium, toxic effects, etc...), (iii) the particular combination of a specific signal sequence with a specific heterologous polypeptide (not all combinations give similar results, incompatibility, etc...), (iv) the production of a polypeptide in one species (such as the native one) cannot provide any expectation in a different (heterologous) system, etc...

However, the board fails to see in the wording of the claims of the request any requirement for a specific yield or level of secretion. The claimed subject matter comprises processes for producing any heterologous polypeptide under any (culture) conditions and using any filamentous fungus as a host, even if they are worse or result in a lower production and secretion of this polypeptide than using the native filamentous fungus as a host and/or under optimal conditions. In other words, the expression and secretion of **any** (low, minimum) amount of this heterologous polypeptide would already fulfil the expectations of the skilled person. The board considers that, whereas a reasonable

expectation of success is notoriously less likely to exist when a specific technical improvement is intended to be achieved, if what is looked for are only alternatives showing more or less the same effect as in the closest prior art, then the skilled person will follow up hints in the prior art which suggest already some likelihood of success in achieving this unambitious aim: success need not be certain (cf eg T 338/00 of 6 November 2002, see point 10 of the reasons).

11. In the present case, the board considers that the skilled person would have had more than a reasonable expectation of success. In particular for the following reasons:

(i) as stated in point 9 above, systems for transforming and expressing polypeptides in filamentous fungi were well-known and available to the skilled person (cf documents D12 and D15). These systems had been further improved for obtaining higher transformation frequencies (cf documents D16 and D23).

(ii) these transformation systems had been successfully used for expressing functionally active heterologous polypeptides in several fungi hosts. The expressed polypeptides were heterologous not only at the level of species (A. nidulans amdS gene in A. niger, cf document D13) but at the level of genera as well (Neurospora crassa pyr4 gene in A. nidulans, cf document D12).

(iii) signal sequences of genes from several filamentous fungi had been shown to be functionally active in non-filamentous fungi hosts such as yeast (Aspergillus glucoamylase in document D7; Trichoderma CBHI cellulase in document D47).

To the extent that the suggested host (filamentous fungi) is more closely related to the source of the gene and signal sequence (T. reesei CBHI cellulase), it was reasonable to expect that the system suggested in document D47 (expression and secretion of T. reesei CBHI cellulase in filamentous fungi) would be even more efficient than the one actually disclosed in this prior art (yeast). Even if a lower yield than the one obtained with the native filamentous fungus (Trichoderma reesei) could be expected when using other filamentous fungi as hosts (but not necessarily), this (yield) feature cannot be taken into account for assessing the inventive step (point 10 above).

12. In view of the foregoing, the board concludes that starting from document D47, the person skilled in the art would have arrived at a method falling within the scope of claim 3 without exercising any inventive activity.

13. A similar argumentation applies to the subject matter of claim 2 of this request. The subject matter of this claim concerns the expression and secretion of a Humicola or a Mucor polypeptide using its native signal and Aspergillus as the filamentous fungus host. As stated in point 11(ii) above, the successful expression of a gene from a filamentous fungus in a different filamentous species as a host was already known in the

prior art (Neurospora crassa pyr4 gene in Aspergillus nidulans, document D12) and knowing that signal sequences from filamentous fungi were functionally active in unrelated non-filamentous fungi hosts (cf documents D7 and D47) (point 11(iii) above), it was reasonable to expect that signal sequences from filamentous fungi (such as from Humicola and Mucor) would generally function in more closely related filamentous fungi species (such as Aspergillus). In the absence of any technical evidence showing that for the particular selection of genes and hosts of claim 2 an unexpected advantage or technical improvement is achieved (point 10 above), the board fails to see which is the actual inventive contribution to the art of the subject matter of this claim and thus no inventive step can be acknowledged.

14. For all these reasons, the board considers that the subject matter of claims 2 and 3 is not inventive and thus, this request, which comprises them, does not fulfil the requirements of Article 56 EPC.

Order

For these reasons it is decided that:

The appeal is dismissed.

The Registrar: The Chairman:

A. Wolinski L. Galligani