Internal distribution code:
(A) [ ] Publication in OJ
(B) [ ] To Chairmen and Members
(C) [x] To Chairmen
(D) [ ] No distribution

Datasheet for the decision of 23 September 2009

Case Number: T 0458/07 - 3.3.04

Application Number: 99921610.4

Publication Number: 1075488

IPC: C07K 1/18

Language of the proceedings: EN

Title of invention:
Protein purification by ion exchange chromatography

Patentee:
Genentech, Inc.

Opponent:
Novo Nordisk A/S

Headword:
Ion exchange chromatography method/GENENTECH

Relevant legal provisions:
-

Relevant legal provisions (EPC 1973):
EPC Art. 54, 56, 83, 123(2), 111(1), 114(2)
EPC R. 27(1)(e)

Keyword:
"Added subject-matter (no)"
"Sufficiency of disclosure, novelty, inventive step (yes)"

Decisions cited:
G 0009/91, T 0939/92, T 0390/07

Catchword:
-

Europäisches Patentamt | European Patent Office | Office européen des brevets

Beschwerdekammern | Boards of Appeal | Chambres de recours

Case Number: T 0458/07 - 3.3.04

D E C I S I O N
of the Technical Board of Appeal 3.3.04
of 23 September 2009

Appellant I: (Patent Proprietor)
Genetech, Inc.
1 DNA Way
South San Francisco CA 94080-4990 (US)

Representative:
King, Hilary
Mewburn Ellis LLP
33 Gutter Lane
London EC2V 8AS (GB)

Appellant II: (Opponent)
Novo Nordisk A/S
Novo Allee
DK-2880 Bagsvaerd (DK)

Representative:
Ford, Hazel
J.A. Kemp & Co.
14 South Square
Gray's Inn
London WC1R 5JJ (GB)

Decision under appeal:
Interlocutory decision of the Opposition Division of the European Patent Office posted 11 January 2007 concerning maintenance of European patent No. 1075488 in amended form.

Composition of the Board:

Chair: U. Kinkeldey
Members: G. Alt
D. S. Rogers

Summary of Facts and Submissions

I. Both, the patent proprietor (hereinafter "appellant I") and the opponent (hereinafter "appellant II") have appealed against the decision of the opposition division according to which European patent EP 1 075 488, entitled "Protein purification by ion chromatography" could be maintained in amended form pursuant to Article 102(3) EPC 1973.

II. The patent had been granted with 22 claims.

The two independent claims, claims 1 and 22, read:

"1. A method for purifying a polypeptide from a composition comprising the polypeptide and a contaminant, which method comprises the following steps performed sequentially:

(a) binding the polypeptide to an ion exchange material using a loading buffer, wherein the loading buffer is at a first conductivity and pH;
(b) washing the ion exchange material with an intermediate buffer at a second conductivity and/or pH so as to elute the contaminant from the ion exchange material;
(c) washing the ion exchange material with a wash buffer which is at a third conductivity and/or pH, wherein the change in conductivity and/or pH from the intermediate buffer to the wash buffer is in an opposite direction to the change in conductivity and/or pH from the loading buffer to the intermediate buffer; and

(d) washing the ion exchange material with an elution buffer at a fourth conductivity and/or pH so as to elute the polypeptide from the ion exchange material.

22. A method for purifying a polypeptide from a composition comprising the polypeptide and a contaminant, which method comprises the following steps performed sequentially:
(a) binding the polypeptide to a cation exchange material using a loading buffer, wherein the loading buffer is at a first conductivity and pH;
(b) washing the cation exchange material with an intermediate buffer at a second conductivity and/or pH which is greater than that of the loading buffer so as to elute the contaminant from the ion exchange material;
(c) washing the cation exchange material with a wash buffer which is at a third conductivity and/or pH which is less than that of the intermediate buffer; and
(d) washing the cation exchange material with an elution buffer at a fourth conductivity and/or pH which is greater than that of the intermediate buffer so as to elute the polypeptide from the ion exchange material.

III. The opposition was based on Article 100(a) EPC, lack of novelty and lack of inventive step, and on Article 100(b) EPC, insufficiency of disclosure.

IV. The present decision refers to the following documents:

D1 WO 96/40883

D6 WO 94/22905

D7 EP 289 129

D9 Declaration by Dr Lester dated 8 November 2006

D10 Blood, vol. 76, no. 12, 1990, pages 2546-2555, Grinnell, B.W. et al.

D11 Declaration by Prof. Hearn dated 16 May 2007

D12 Declaration by Dr Staby dated 21 May 2007

D13 Biotechnology and Bioengineering, vol. 42, 1993, pages 1086-1090, Ming, F. et al.

D14 Chromatographia, vol. 28, 1989, pages 170-178, Levison, P.R. et al.

D15 Biochimica et Biophysica Acta, vol. 952, 1988, pages 201-207, Cole, T.-Ch. et al.

D16 Biochimie, vol. 70, 1988, pages 227-236, Sanchez-Bernal, C. et al.

D17 Declaration by Dr Krarup dated 21 May 2007

D18 Declaration of Dr Ahmadian dated 21 May 2007

D27 WO 96 35718

D29 Declaration of Dr Dowd dated 14 August 2008

D32 Second Declaration of Dr Ahmadian dated 27 July 2009

D34 The Journal of Biological Chemistry, vol. 99, no. 3, 1933, pages 741-753, Wintersteiner, O. and Abramson, H.A.

V. The opposition division held that due to an error in the disclosure the technical information disclosed in Example 4 of document D7 did not constitute prior art and was therefore not relevant to the novelty of the subject-matter of claim 1 of the main request (i.e. the claims as granted). This subject-matter and that of claim 1 of auxiliary requests 1 and 2, respectively, was found to lack novelty over the disclosure in each of documents D1 and D6. The claims of the third auxiliary request were held to comply with the requirements of the EPC.

VI. With the statement of grounds of appeal, appellant II submitted documents D10 to D23, among them declarations of Prof. Hearn (D11), Dr Staby (D12), Dr Krarup (D17) and Dr Ahmadian (D18). With a further submission two further documents and a second declaration of Dr Ahmadian (D32) were filed. Appellant I submitted in all seven new documents, among them the declaration of Dr Dowd (D29).

VII. On 25 February 2008, i.e. during the appeal proceedings, observations by a third party pursuant to Article 115 EPC were filed together with document D27.

VIII. In a communication the board informed the parties about its intention to admit document D27 into the procedure due to its relevance for the novelty of some of the

claimed subject-matter and invited the parties to comment.

IX. In a first response to the third party's observations appellant I requested remittal of the case to the opposition division for consideration of document D27. In a second response, appellant I filed a new main request and auxiliary requests I to IV. In a further submission appellant I filed a replacement auxiliary request II and a new auxiliary request V.

X. Oral proceedings were summoned with a communication dated 19 February 2009 and were held on 23 September 2009. At the oral proceedings appellant I filed a new main request corresponding to the previous main request with the exception of amended claim references in claims 12 and 15.

The main request encompasses three independent claims.

Independent claims 1 and 2 read:

"1. A method for purifying a polypeptide from a composition comprising the polypeptide and a contaminant, which method comprises the following steps performed sequentially:

(a) binding the polypeptide to an ion exchange material using a loading buffer, wherein the loading buffer is at a first conductivity;
(b) washing the ion exchange material with an intermediate buffer at a second conductivity which is greater than the conductivity of the loading buffer so

as to elute the contaminant from the ion exchange material;
(c) washing the ion exchange material with a wash buffer which is at a third conductivity which is less than the conductivity of the intermediate buffer; and
(d) washing the ion exchange material with an elution buffer at a fourth conductivity which is greater than the conductivity of the intermediate buffer so as to elute the polypeptide from the ion exchange material,

wherein elution of the contaminant and of the polypeptide is achieved by modifying the conductivity of the intermediate buffer and of the elution buffer, respectively.

2. A method for purifying a polypeptide from a composition comprising the polypeptide and a contaminant, wherein the polypeptide is an antibody, which method comprises the following steps performed sequentially:

(a) binding the polypeptide to an ion exchange material using a loading buffer, wherein the loading buffer is at a first conductivity and pH;
(b) washing the ion exchange material with an intermediate buffer at a second conductivity which is greater than the conductivity of the loading buffer and/or at a second pH so as to elute the contaminant from the ion exchange material;
(c) washing the ion exchange material with a wash buffer which is at a third conductivity which is less than the conductivity of the intermediate buffer and/or at a third pH, wherein the change in conductivity and/or pH from the intermediate buffer to the wash

buffer is in an opposite direction to the change in conductivity and/or pH from the loading buffer to the intermediate buffer; and
(d) washing the ion exchange material with an elution buffer at a fourth conductivity which is greater than the conductivity of the intermediate buffer and/or at a fourth pH so as to elute the polypeptide from the ion exchange material.

The third independent claim of this main request, claim 22, was the same as claim 22 as granted (see above section II).

XI. Appellant I's final requests at the oral proceedings were that the decision under appeal be set aside and that the patent be maintained upon the basis of claims 1-22 of the main request or on the basis of one of five auxiliary requests, that documents D10 to D23 be not admitted into the proceedings and that the case be remitted to the department of first instance for consideration of document D27.

Appellant II requested the decision under appeal be set aside and that the patent be revoked.

XII. Appellant II's (opponent's) arguments, as far as they are relevant to the present decision, may be summarized as follows:

Admission of documents D10 to D23

Documents D10 to D23 had been filed at the earliest possible point in time during the appeal proceedings and therefore could be properly dealt with by the

parties and the board. Thus, they should be admitted into the proceedings.

Article 123(2) EPC

Amended claim 1 related to both anion and cation chromatography, but none of the passages in the application as filed indicated by appellant I was a clear and unambiguous basis for the claimed conductivity pattern in relation to anion chromatography. In particular, the statement on page 17, lines 35 to 36 that the changes in conductivity were "generally" as described for cation chromatography could also mean that they could be different therefrom. Moreover, claims 4 to 6 as filed, which recited the wording now used for the definition of the buffer pattern in claim 1, referred to claim 2 as filed which related only to cation exchange chromatography. Thus, the requirements of Article 123(2) EPC were not fulfilled.

Sufficiency of disclosure

In the present case one example was not a sufficient disclosure to support the broad claim.

It was even not sure whether the only example properly represented the claimed subject-matter. For example, the conductivities of the buffers were not disclosed in the example; there was a further wash step between the loading and the wash with the intermediate buffer which was not a feature of the method as claimed. Thus, the example did not clearly disclose whether the claimed effect was due to the buffer changes. Moreover, it did

even not disclose the intended effect, i.e. purification of the protein from all contaminants because, in fact, only one of several contaminants could be removed.

Declarations D17 and D18 demonstrated that the invention could not be carried out over the whole scope of the claim with undue burden. Both declarants, Dr Krarup and Dr Ahmadian, had worked according to the instructions in the patent, yet Dr Krarup could not purify human insulin from its deamidated variant on an anion chromatography exchanger (declaration D17) and Dr Ahmadian could not remove contaminants from a preparation of Factor VII (declaration D18).

Novelty

Documents D7, D10, D27

Document D7

In view of the conductivities stated in the declaration by Dr Lester (document D9) the process disclosed in Example 4 of document D7 had all the features recited in claim 1. As submitted in the declarations D11 and D12 by Prof. Hearn and Dr Staby, given the many variations in which ion chromatography could be conducted, the skilled person would not have recognized any error in the description of the buffer compositions in Example 4. For example, it was neither unusual to use three buffers with the same composition during one chromatography run, see for example the Equilibration buffer 2 and wash buffers 1 and 3 in Table 1 of the patent, nor to use Tris at high concentrations in

chromatography buffers nor to use a single equilibration buffer (see documents D13 to D16). Finally, patent documents were always made with a high level of accuracy so that errors would not be expected. Thus, the disclosure in document D7 anticipated the subject-matter of claim 1.

The disclosure in document D8, the corresponding US family member, was the same as in document D7. As to document D24 it was not known whether the method disclosed therein was even intended to be the same as in document D7.

Document D10

Document D10 disclosed a process for the purification of human protein S (HPS). According to the declaration D18 the conductivities of the buffers used in document D10 were between 14.73 mS/cm and 14.77 mS/cm (determined by three independent measurements) for the loading buffer, between 17.92 mS/cm and 17.96 mS/cm (four independent measurements) for wash buffer 1 (corresponding to the "intermediate buffer" in the claimed process), between 17.62 mS/cm and 17.75 mS/cm (four independent measurements) for wash buffer 2 (corresponding to the "wash buffer" in the claimed process) and between 18.19 mS/cm and 18.26 mS/cm (four independent measurements) for the elution buffer 1 (corresponding to the "elution buffer" in the claimed process).

Although the difference in the conductivity between wash buffer 1 and 2, i.e. between the intermediate and wash buffer in the terms of the patent, was small, it

could not be neglected that the conductivity of the wash buffer 2 was lower.

The lower conductivity of the wash buffer 2 could not be regarded as a measurement irregularity, but was due to the lack of EDTA in that buffer compared to wash buffer 1.

It did not matter that elution according to the process disclosed in document D10 was achieved by a conformational change of the protein with the help of calcium ions, since, nevertheless, the elution buffer had a higher conductivity than the wash buffer.

Thus, the process disclosed in document D10 was novelty-destroying for the subject-matter of claim 1.

Document D27

Request for remittal to the department of first instance for consideration of document D27

Document D27 was highly relevant for the novelty of the claimed subject-matter. It had been filed by the third party at such an early stage during the appeal proceedings that it could be properly dealt with by the parties and the board. Therefore, the case should not be remitted to the department of first instance for consideration of document D27.

Substantive issues

Document D27 disclosed in Example 6 a process for the purification of erythropoietin. On the basis of the

conductivities as disclosed in the third party's submission, which were 1.8 mS/cm for the loading buffer, 2.7 mS/cm for the buffer corresponding to the "intermediate buffer" in the patent, 1.8 mS/cm for the buffer corresponding to the "wash buffer" in the patent and 9.6 mS/cm for the elution buffer, the process disclosed in Example 6 of document D27 was novelty-destroying for the subject-matter of claim 1.

I*nventive step*

Each of documents D1, D6, D7 and D10 disclosed an ion chromatography process wherein the conductivity of the wash buffer was lower than the conductivity of the intermediate buffer and the conductivity of the elution buffer was greater than that of the intermediate buffer. Thus, the claimed subject-matter was obvious in view of each of these documents.

An inventive step had to be denied also because not substantially all embodiments of the claims achieved the intended effect.

XIII. Appellant I's (the patent proprietor's) arguments, as far as they are relevant to the present decision, may be summarized as follows:

Admission of documents D10 to D23

An appeal should be based on the facts and evidence that were available to the department of first instance. Therefore, documents D10 to D23, which were filed only at the appeal stage should not be admitted.

Article 123(2) EPC

The statement on page 17, lines 35 and 36 clearly meant that the general outline of conductivity changes disclosed with regard to cation chromatography in the passages above this statement also applied to anion chromatography. This meaning was supported by the passage following the statement emphasizing that the relation of the buffers relative to each other was different when elution was achieved by a change in pH. Thus, amended claim 1 had an explicit basis in the application as filed.

Sufficiency of disclosure

When considering the disclosure in the patent as a whole in the light of the common general knowledge, the skilled person could carry out the claimed invention.

The assays disclosed in declarations D18 and D17 were not suited to show that the claimed method could not be carried out without undue burden over the whole scope of the claim.

Novelty

Documents D7, D10, D27

Document D7

The skilled person would immediately consider it odd that three differently named buffers - "Anion-Exchange Column Equilibration Buffer #1*", "Anion-Exchange Column Equilibration Buffer #2*" and "Anion-Exchange

Column Wash Buffer #1* - and which were used in series, had the same composition.

A Tris concentration of 250mM in "Anion-Exchange Column Equilibration Buffer #2*" also made no sense when regarding "Anion-Exchange Column Wash Buffer #1*" and "Anion-Exchange Column Wash Buffer #2*", the composition of "Anion-Exchange Column Wash Buffer #1*" being defined as "Same as Anion-Exchange Column Equilibration Buffer #2*". Since the purpose of the second wash was to remove Triton before elution of the desired protein, the Tris concentration in wash buffer 1 and 2 should be the same, i.e. 25mM as in wash buffer 2.

The disclosure in documents D8 and D24 would have confirmed the skilled person's view.

Thus, in fact, the conductivity pattern of the buffers disclosed in document D7 were not as claimed and therefore the document did not destroy the novelty of the subject-matter of claim 1.

Document D10

In the process according to document D10 elution of the desired protein was achieved by running an ion chromatography column in a pseudo-affinity mode, i.e. calcium ions in the elution buffer bind to human protein S (HPS), induce a conformational change of the polypeptide and thereby release it from the column. Thus, since this is not an elution caused by the change in the conductivity of the elution buffer, document D10 does not disclose the feature in claim 1 that the

elution of the polypeptide was achieved by modifying the conductivity of the elution buffer. Moreover, the feature in step b of claim 1, that the contaminant elutes by virtue of the intermediate buffer was not disclosed in the document. Hence document D10 was not relevant to the novelty of the subject-matter of claim 1.

Document D27

Request for remittal to the department of first instance for consideration of document D27

As stated in decision G 9/91 the purpose of an appeal was to give the losing party the possibility of challenging the decision of the first instance. The function of the boards in the appeal proceedings was not to decide upon questions raised for the first time during these appeal proceedings. Since document D27 had been filed by the third party only at the appeal stage, remittal to the department of first instance for the examination of document D27 was thus appropriate.

Substantive issues

It was stated in the declaration by Dr Dowd (document D29, Table 1) that the conductivity value for the intermediate buffer was not 2.7 mS/cm as submitted by the third party, but that it was between 0.92 mS/cm and 1.04 mS/cm. Thus, there was no increase of the conductivity between the loading buffer and the intermediate buffer and therefore, document D27 was not novelty-destroying for the subject-matter of claim 1.

Inventive step

None of documents D1, D6, D7 or D10 taught that the decrease of the conductivity of the wash buffer relative to the intermediate buffer and the increase of the conductivity of the elution buffer relative to the intermediate buffer would result in a better separation of a given protein from a contaminant.

XIV. At the end of the oral proceedings the board announced its decision.

Reasons for the decision

Admission of documents D10 to D23 and documents D24 to D34 into the proceedings

1. Pursuant to Article 114(2) EPC a board may disregard facts and evidence which are not submitted in due time by the parties. Thus, it follows from this provision that the parties can file new matter during the appeal proceedings. This is confirmed by Article 13(1) of the Rules of the Procedure of the Boards of Appeal which gives the board a discretion to decide upon amendments made to a party's case even after the statement of the grounds of appeal has been filed. Thus, the appeal proceedings according the EPC are not restricted to the consideration of the material which had already been presented at the first instance.

2. In view of Article 114(2) EPC the non-admission of facts and evidence is at a board's discretion. In exercising this discretion the parties' right to be heard and to a fair conduct of the proceedings is to be

taken into account, as well as the public's interest in a speedy outcome of the proceedings and the existence of valid patents. Criteria considered by the boards have been thus, inter alia, the complexity of the new material, the point in time during the proceedings of its filing, the reason for its filing and its relevance.

3. Documents D10 to D23 were filed with appellant II's statement of the grounds for appeal in May 2007, thus at the very beginning of the appeal proceedings. Taking into account that oral proceedings took place in September 2009, the parties and the board had sufficient time to consider the document so that both parties' right to be heard pursuant to Article 113(1) EPC is safeguarded without a remittal.

The board has therefore considered it appropriate not to disregard documents D10 to D23.

4. Documents D24 to D31 were filed in August 2008, i.e. more than one year before oral proceedings took place, i.e. in the board's view long enough for their proper consideration. Documents D32 to D34 were filed one month before the oral proceedings. Document D32, i.e. the second declaration of Dr Ahmadian, is a direct response to appellant I's criticism with regard to Dr Ahamdian's first declaration, document D18. Hence the document is a reaction on a procedural development. So are documents D33 and D34 which were submitted in relation to the declaration by Dr Krarup (D17) in order to show the pI value for human insulin. Since the contents of the documents are not so complex that they could not be dealt with during the oral proceedings, the board considers it appropriate to exercise its

discretion such as to admit documents D24 to D31 and documents D32 to D34 into the proceedings, the more so, since none of parties has requested not to admit any of these documents.

Article 123(2) EPC

5. Claim 1 relates to a method for purifying a polypeptide from a composition comprising the polypeptide and a contaminant by ion exchange chromatography. There are only two types of ion exchange chromatography, namely anion and cation chromatography.

6. Appellant II argues that, the conductivity changes of the buffers as set out in claim 1 are not clearly and unambiguously derivable from the application as filed as far as claim 1 relates to the purification by anion chromatography.

7. In a passage starting on page 16, line 35 and continued on page 17, up to line 33 of the application as filed, it is explained how the conductivity and/or the pH of the different chromatography buffers is/are changed relative to each other according to the invention. The passage starts as follows: "With particular reference to Figure 1, which shows exemplary steps to be performed where a cation exchange resin is used, the pH and/or conductivity of each buffer is/are increased relative to the preceding buffer, except for the wash buffer where the conductivity and/or pH is/are less than the conductivity and/or pH of the preceding intermediate buffer." This summary is followed by a more detailed elaboration of the changes in

conductivity and pH of the buffers for the purification of a polypeptide with cation chromatography.

In particular it is also derivable from the cited passage that the specific conditions disclosed in Figure 1, i.e. inter alia the specific salt concentrations and pH values of the different buffers, are to be considered only as an example of the general teaching ("which shows exemplary steps").

8. Immediately after the end of the passage concerning cation chromatography on page 17, line 33 a new paragraph starts in line 34: "In an alternative embodiment, the ion exchange material comprises an anion exchange resin. **This embodiment of the invention is depicted in Figure 2 herein. As illustrated in this figure, the changes in conductivity are generally as described above with respect to a cation exchange resin. However, the direction of change in pH is different for an anion exchange resin.** For example, if elution of the contaminant (s) and polypeptide are to be achieved by altering pH, the loading buffer has a first pH and the pH is decreased in the intermediate buffer so as to elute the contaminant or contaminants. In the third step, the column is washed/re equilibrated with the wash buffer and the change in conductivity or pH, or both, is in the opposite direction to that of the previous step. Hence, the pH may be increased in the wash buffer, compared to the intermediate buffer." (emphasis added).

9. In the board's view, the skilled person would clearly and unambiguously derive from this whole passage that for the separation of protein and contaminants by anion

chromatography the relative conductivities of the buffers are the same as those for cation chromatography. The skilled person's understanding would in the board's view be particularly confirmed by the explicit statement that the conditions are different, if elution was to be achieved by pH change.

Even if it is assumed for the sake of argument that the skilled person had considered the term "generally" to mean that the conditions may also be different from that disclosed for cation chromatography, it remains true that the application as filed also discloses a situation where the conditions are the same as for cation chromatography.

10. There is no part of the application that would change the skilled person's understanding derived from the passage cited above. In particular, the skilled person would consider that the teaching with regard to anion chromatography is restricted to the specific values of the Figure 2 in view of the express statement in the passage cited above that the figure "illustrates" this embodiment of the invention.

11. Moreover, the statement on page 16, lines 33 and 34 of the application as filed, i.e. that "[t]he various buffers used for the chromatography depend, for example, on whether a cation or anion exchange resin is employed. This is shown more clearly in the flow diagrams of Figures 1 and 2." would not call into doubt the clear and unambiguous teaching of the passage referred to above.

This is so because the skilled person would derive from the application as filed that the essence of the invention is a particular scheme of changing the conductivity or pH of the buffers relative to each other and would therefore not interpret the reference to "various buffers used" in a way which is in contradiction to the overall teaching in the patent in suit. Rather on the basis of common general knowledge the skilled person would consider that the variation of the buffer in this context refers to the composition of chromatography buffers as far as, for example, its constituents or the salt concentration are concerned.

12. Also the argument that the wording now used in claim 1 for describing the relative buffer conductivities is explicitly used in the claims as filed only with regard to cation chromatography does not convince the board of the absence of a basis in the application as filed for claim 1 as far as anion chromatography is concerned, firstly, because of the clear and unambiguous disclosure in the passage cited above in point 8 and secondly, because claim 1 as filed did not exclude the buffer pattern now claimed for anion chromatography.

13. Thus, the subject-matter of amended claim 1 does not extend beyond the content of the application as filed.

14. Independent Claim 2 is a combination of claims 1 and 15 as filed. Apart from adapted claim references, dependent claims 3 to 9 and 11 to 21 correspond to claims 2 to 8, 10 to 14 and 16 to 21 as filed, respectively. Claim 10 has a basis in the description on page 18, line 20. Claim 22 is the same as claim 23 as filed. Appellant II did not raise objections

pursuant to Article 123(2) EPC with regard to these claims.

15. The requirements of Article 123(2) EPC are fulfilled.

Articles 123(3) and 84 EPC

16. Claim 1 is limited to the alternative in claim 1 as granted that chromatography is conducted by changes in buffer conductivity. Claim 2 corresponds to claim 1 as granted with the restriction that the polypeptide is an antibody. Dependent claims 3 to 21 correspond to dependent claims 3 to 16 and 18 to 21 as granted except that the claim dependencies are adapted. Claim 22 is the same as claim 22 as granted. Thus, the scope of the claims is not extended vis-à-vis the claims as granted. No objection was raised by appellant II in this respect.

Moreover, the board has no objections with regard to Article 84 EPC and none were invoked by appellant II.

17. The requirements of Articles 84 and 123(3) EPC are fulfilled.

Sufficiency of disclosure

18. Appellant II argues that the one example in the patent is not enough for that the disclosure in the patent could be regarded as sufficient. This was the more so, since the example apparently does not correctly represent the claimed subject-matter because the conductivity of the loading buffer is not stated, an additional wash step is carried out and not all contaminants are removed.

19. Rule 27(1)(e) EPC 1973 stipulates that the description shall "describe in detail at least one way of carrying out the invention claimed **using examples where appropriate** and referring to the drawings, if any" (emphasis added). Thus, it follows from this rule that there is no mandatory requirement for an example under the EPC. Hence, even the absence of an appropriate example would not be a reason for denying sufficiency of disclosure.

20. The sufficiency of disclosure with regard to claimed subject-matter is judged on the basis of the whole disclosure in a patent and not only on that given by the examples. Furthermore, the skilled person's knowledge at the time of priority of the patent must be taken into account (Case Law of the Boards of Appeal of the European Patent Office, 5th edition 2006; II. A. 1., first paragraph; II.A.2.a), first paragraph).

21. At the priority date of the patent ion chromatography was a well-established technique for the purification of proteins. Ion chromatography separates proteins according to their charge. The separation is based on the reversible interaction of the protein to an oppositely charged chromatography material. An anion exchange resin has a positively charged surface. A cation exchange resin has negatively charged groups.

Initially, counter ions, i.e. ions of opposite charge present in the equilibration buffer are bound to the charged immobile functional groups of the chromatography resin. When a sample is added to the column, proteins with opposite charge to the resin

displace the counter ions and are absorbed onto the column. Thus, negatively charged proteins bind to the anion exchange resin and positively charged proteins bind to the cation exchange resin.

Proteins bind to the resin when the conductivity of the buffer is low. The conductivity of the buffer is strongly dependent on the salt concentration in the buffer.

Differential elution of the bound proteins can be achieved by altering the conductivity (i.e. the salt concentration) of the buffer or by altering the pH. Elution can be stepwise or in a gradient. When elution is due to a change in the conductivity, the principle is that those proteins that due to their charge are less strongly associated with the resin are displaced from the column at lower conductivities (i.e. salt concentration), while the more strongly associated proteins are eluted at higher conductivities.

At the priority date the skilled person was also aware of a wide variety of combinations of constituents to be used for the chromatography buffers and that the type of resin and the buffers used depended on a given protein. Thus, the skilled person knew that fine-tuning of the chromatography process was necessary for each protein-contaminant-situation, but that this could be achieved on the basis of the well-known physico-chemical principles on which ion-chromatography relies.

22. Some of the above-mentioned knowledge about ion chromatography is summarized in paragraph [0005] of the patent in suit. The skilled person also learns from

this paragraph that the claimed ion chromatography method is meant as an improvement of the "standard" way of conducting ion chromatography (for a "non-standard" way, see for example point 51 below),i.e. where separation of contaminant and protein are achieved by **progressive increase** of the salt concentration in the buffer. Moreover, the skilled person would recognize in view of the claims stating that the contaminant is eluted before the desired protein (see in the preamble "performed sequentially" and steps b) and d)), that the claimed method is an improvement of such ion chromatography situations where the contaminant elutes **before** the protein.

23. Given all this knowledge, in the board's view, the skilled person would not have any difficulty in performing the claimed method on the basis of the disclosure in the patent and this would even be so if the example would not reflect the claimed method.

24. The board notes however in passing that it considers the example to describe a situation falling under the method of claim 1.

- In particular, the conductivity of the loading buffer is indicated in Table 1.

- As regards the additional wash step included in the example the board considers that its presence in the example and absence in the claim could only give rise to an objection pursuant to Article 83 EPC, if this step was an essential feature of the invention. If this was so, the issue would rather

have to be dealt with under Article 84 EPC than under Article 83 EPC.

– Finally, the failure to demonstrate in the example the removal of all contaminants is in the board's view also no reason for considering that the example does not reflect the invention as claimed. Firstly, the claims expressly relate to "purifying a polypeptide from a composition comprising the polypeptide and **a** contaminant and to the elution of "**the** contaminant" (see preamble and step b); emphasis added). Thus, removal of all impurities is not required by the claim. Secondly, in view of the general principles on which ion chromatography is based (see above point 21) and given the differing charges of the variants it would not even be expected by the skilled person that it would be possible to remove all impurities by a single round of chromatography.

25. In a second line of argument appellant II submits that the invention cannot be carried out over the whole breadth of the claim without undue burden. Declarations D17 (Dr Krarup) and D18 (Dr Ahmadian) are the evidence relied on.

26. Dr Ahmadian's (D18) experiments relate to the purification of factor VII (FVII) from its degradation and cleavage products and its oxidized forms (paragraph 2.2). Apart for the control experiments the conductivity pattern of the buffers was adjusted according to the disclosure in the patent (paragraph 2.1). Experiments A1, A2, A4 and A5 were conducted according to the principles of "classical" ion

chromatography, i.e. by progressive increase in the salt concentration in the buffers. Experiments A3, A6 and A7 were conducted according to the method of the patent (see Table 1 and "Results and Interpretation"). It is reported that in **none** of the experiments reduction of contaminants in eluate was seen (paragraph 4.1).

27. The method of the invention is to be seen as an improvement of the "typical" ion chromatography in that it gives a better separation of contaminants from desired protein (see for example paragraphs [0005] and [0006] of the patent; see also point 22 above). Thus, an improvement is only achievable with the method of the invention, if a given protein-contaminant pair is also separable by the "classical" method. This is reflected by the specific example in the patent that the amount of impurities was 25% when using the "normal" method, but that it could be reduced to 13% or less when using the method of the patent (paragraph [0107]).

28. Experiments A1, A2, A4 and A5 in document D18 demonstrate that FVII is not separable from contaminants under the selected circumstances even by the "standard" ion chromatography method. Hence, it is not surprising that there is no separation when chromatography is conducted according to the concept of the patent. Thus, the experiments disclosed in document D18 do not prove that the method disclosed in the patent does not achieve the expected result.

29. Dr Krarup's experiments in document D17 relate to the separation of human insulin from its deamidated variant

on an anion exchanger at pH 8 (paragraph 2.2). He found that there was essentially no purification (paragraph 4.1). Appellant II argues that, since the patent expressly taught that the method of the invention was suited to purify a protein from a deamidated variant, generally and in the specific example, the experiments in document D17 demonstrated that the claimed method did not actually work across the whole claimed scope.

30. However, as stated above in point 22 the teaching of the patent is applicable to situations where the contaminant elutes before the desired protein from the ion chromatography column. In view of known physico-chemical principles, the skilled person would be aware that this is not a mandatory constellation, in cases where the contaminant is a deamidated variant of the desired protein. The reason is as follows: the charge of a protein under given pH conditions depends on the isoelectric point (pI) of that protein. The pI is the pH at which the negative and positive charges of the protein are equal. For example, human insulin used in document D17 has a pI of approximately 5.3 to 5.8 (see document D34).

The lack of at least an amino group has the consequence that a deamidated variant of a given compound always has a lower pI than the compound itself. This is also apparent from the example in the patent, disclosing in paragraph [0087] that the HER-2 antibody has a pI of 8.87 while the singly-deamidated variant thereof has a pI of 8.79. As a result of the lower pI of the de-amidated variant in relation to the amidated parent compound, the deamidated compound is more negatively charged at a pH above its pI than its amidated

counterpart. In contrast, at a pH below the pI the amidated compound is more negatively charged than the deamidated variant

The consequence under the conditions selected by Dr Krarup, i.e. separation of human insulin having a pI between 5.3 and 5.8 from its deamidated variant on an anion exchanger at pH 8 is that the deamidated human insulin variant has more negative charges than the amidated variant and therefore binds more tightly to the positively charged anion exchange resin than the amidated molecule. It may therefore be concluded on the basis of these theoretical observations that upon increase of the salt concentration in the buffer, i.e. upon increase of the conductivity of the buffer, the deamidated insulin variant is likely to elute **after** the parent molecule. This may explain why the content of deamidated variant in fractions 6-8 is low (see Table 4 of document D17, samples denoted "Front flank").

These observations may also explain why in the experiments of document D17 even with the standard approach, i.e. a single wash followed by elution, separation of human insulin from its desamido-variant was not achieved (sample "HW07-013-05"; paragraph 3.5; chromatogram in Annex 3 to document D17).

In contrast, under the conditions of the specific example of the patent, separation of HER-2 antibody having a pI of 8.87 from its deamidated variant having a pI of 8.79 (see paragraph [0087] of the patent) by cation chromatography at pH 5.6 (see paragraphs [0098] and [0099] of the patent), the deamidated variant is more negatively charged, will therefore bind less

tightly to the negatively charged cation resin than the amidated parent antibody and therefore elute **before** it.

31. Hence, the board concludes that human insulin could not be separated from its deamidated variant under the "standard" ion chromatography conditions. Consequently, for the reasons given above given in relation to document D18, also the experiments in document D17 do not prove that the method of the patent does not work.

32. The board concludes that no case has been made that the invention cannot be carried out over the whole breadth claimed without undue burden.

33. The requirements of Article 83 EPC are fulfilled.

Novelty

Documents D7, D10, D27

Document D7

34. Document D7, a European patent application, deals with the purification of protein A from endotoxin (page 3, lines 14 to 16). Purification involves ion chromatography (see bottom of page 3). Document D7 discloses on page 5 buffers that may be used during an anion chromatography purification process. Further details such as the composition of the solution loaded to the column, and the order in which the buffers are used are disclosed in Example 4 and in the last two lines on page 7, respectively.

35. The composition of the buffers is disclosed on page 5 in the following way:

Anion-Exchange Column Equilibration Buffer #1*	0.25 M Tris-HCl pH 8.3 2 mM potassium ethylenediamine-tetraacetic acid (KEDTA) 0.1 mM phenylmethylsulfonyl-fluoride (PMSF) 0.2% TRITON® X-100
Anion-Exchange Column Equilibration Buffer #2*	0.25 M Tris-HCl pH 8.3 2 mM KEDTA 0.1 mM PMSF 0.2% TRITON® X-100
Anion-Exchange Column Wash Buffer #1*	Same as Anion-Exchange Column Equilibration Buffer #2
Anion-Exchange Column Wash Buffer #2*	25 mM Tris-HCl pH 8.3
Anion-Exchange Column Elution Buffer*	25 mM Tris-HCl pH 8.3 150 mM KCl

36. Thus, according to this description the Anion Exchange Column Equilibration Buffers #1* and #2* and the Anion Exchange Column Wash Buffer #1* have an identical composition. Anion Exchange Column Wash Buffer #1* of document D7 corresponds to the "intermediate buffer" and Anion Exchange Column Wash Buffer #1* corresponds to the wash buffer in claim 1 of the patent.

37. The conductivity of the buffers disclosed in document D7 is according to document D9, the declaration by Dr Lester, as follows (using the terms in the patent):

- loading solution: 1.45 mS/cm
- intermediate buffer: 8.78 mS/cm
- wash buffer: 0.9 mS/cm
- elution buffer: 19.7 mS/cm.

Thus, when the disclosure in document D7 are taken at face value, the conductivities of the disclosed buffers fall under the terms of claim 1.

38. Appellant I submits that the skilled person would not have taken the disclosure at issue at face value. He/she would immediately have considered the disclosure of the sequential use of two identically composed equilibration buffers to be wrong, the more so, since they are denoted with different names. Therefore, the Anion Exchange Column Wash Buffer #1*, which is defined by reference to Anion Exchange Column Equilibration Buffer #2* also had to have a composition different from the indicated one.

39. Appellant II argues on the basis of documents D11 and D12 that the protocol in document D7 would have appeared plausible to the skilled person. Firstly, the high concentration of Tris in Anion Exchange Colum Wash Buffer #1* would not strike the skilled person because he/she knows that Tris is an effective buffer, but a poor eluting species and that it can therefore be used over a wide range of concentrations in chromatography buffers. Secondly, it is not unusual that the equilibration and loading buffers had different compositions. Thirdly, it was common at the priority date of the patent to use a single equilibration buffer in order to remove the storage solution and prepare

column for loading, see for example documents D13 to D16.

However, the board is not convinced by any of appellant II's evidence since none of it has a bearing on the question of whether or not the use of two identically composed buffers at two different points during the chromatography process for the same purpose, i.e. for equilibration, would have appeared reasonable to the skilled person. In particular, documents D13 to D16 do not disclose a two-step equilibration, but equilibration by one step. The board notes that a two-step equilibration in the context of anion chromatography is disclosed in Example 6 of document D27 (see points 59 to 65 below). However, the two buffers have a different composition. It is stated in point 6.2 of document D27: "Anschliessend wird die Säule zunächst mit 100mM Na/K-Phosphatpuffer, pH 7,5 and dann mit mindestens 12 SV Äquilibrierpuffer äquilibriert." The equilibration buffer has a concentration of 10 mM Na/K phosphate.

40. Also the further argument that the skilled person would assume that patent documents are always made with a high level of accuracy and that therefore the skilled person would trust the information in the patent document D7 does not convince the board. Typographical and other errors even in patent documents can never be excluded and the board is convinced that also a patent document would be read with technical expertise.

41. Thus, after consideration of the appellants' arguments, the board is more convinced by appellant I's submission and therefore comes to the conclusion that the skilled

person would have considered the sequential use of two identical equilibration buffers which are denoted with different names as a mistake.

42. However, it is not the mandatory consequence of this finding in the present case, that - as submitted by appellant I - the disclosure does not form part of the state of the art. Rather, the subsequent question is what the skilled person would have considered as the intended meaning of this erroneous disclosure.

43. Appellant I argues that, in the light of the overall teaching in document D7 and in view of either of documents D8 or D24, the skilled person would consider that the Tris concentration in Anion Exchange Column Wash Buffer #1* should in fact amount to 25mM.

44. In fact, according to the teaching in document D7 the separation of protein A from endotoxin is not achieved by a change in the conductivities, but by inclusion of a non-ionic detergent such as Triton X100 in the buffers. The detergent supports the dissociation of protein A and endotoxin. Only for elution of protein A is an increase in the conductivity of the buffer necessary. Thus, the skilled person would recognize that there is no need for a change of the conductivities between Anion Exchange Column Wash Buffer #1* and Anion Exchange Column Wash Buffer #2*. Consequently, in the board's view, the skilled person would consider it plausible if the Tris- concentration in the two wash buffers was the same, i.e. 25mM.

Given that this conclusion is reached on the basis of document D7 alone, the issues relating to documents D8

and D24 (see section XIII above) need not be dealt with.

45. According to document D9, the conductivity of the "corrected" Anion Exchange Column Wash Buffer #1 of document D7, i.e. having a Tris concentration of 25 mM, is 1.44 mS/cm. Thus, in the process disclosed in document D7 the conductivity of the intermediate buffer ("Anion Exchange Column Wash Buffer #1") is not greater than the conductivity of the loading buffer as required by steps a) and b) of claim 1. Hence, document D7 does not disclose a process falling under the terms of claim 1.

Document D10

46. Document D10 discloses on page 2547, second column a process for the purification of recombinant human protein S (HPS). The protein preparation is loaded onto an anion-exchange column, the column is washed twice and finally the protein is eluted.

47. According to document D18, the first declaration by Dr Ahmadian, the conductivities of the buffers are as follows (mean values; buffers denoted according to the language of the patent):

48.

- loading buffer: 14.7 mS/cm
- intermediate buffer: 17.9 mS/cm for
- wash buffer: 17.7 mS/cm
- elution buffer: 18.2 mS/cm.

49. The difference in conductivities between the intermediate buffer and wash buffer is small.

Nevertheless, in the board's view, it cannot be regarded as a measurement irregularity because the composition of the two buffers is indeed different, i.e. the composition of the two buffers is identical with the exception that EDTA is omitted from the wash buffer. This explains the difference of the conductivity.

50. Since there is no feature in the claim 1 determining the extent of the decrease of conductivity between the intermediate and the wash buffer, the conductivity pattern of the buffers used in the process disclosed in document D10 falls under the definition of the conductivity pattern in claim 1.

51. Appellant I considered the subject-matter of claim 1 to be novel vis-à-vis the disclosure in document D10 for the following reason:

According to document D10, fully active HPS is eluted from the anion chromatography column in that calcium ions in the elution buffer induce a conformational change in the column-bound protein resulting in its desorption from the column. This way of elution is called "pseudo-affinity" elution in document D10 (page 2552, second column, first full paragraph). In contrast, according to the "classical", conductivity-mediated elution, desorption of the protein from the column is achieved predominantly by a change in the charge of the protein.

52. Appellant I submits that the elution by the pseudo-affinity mode does not fall under the terms of the expression at the end of claim 1 "wherein elution [...] of the polypeptide is achieved by modifying the

conductivity [...] of the elution buffer ...". The expression had to be interpreted such that it only related to elution by a change of the charge of a protein.

53. The board does not agree with this interpretation. In the board's view, the feature cited above does not define the manner in which the protein is eluted from the column, i.e. elution by change of the charge or by change of the conformation of the protein because it is only stated how the buffer is changed for elution, i.e. elution is achieved by modifying the conductivity of the buffer. Elution via the pseudo-affinity route requires the addition of calcium ions to the elution buffer and this addition inevitably changes the conductivity of the buffer. Hence, the feature of claim 1 cited above is disclosed in document D10.

54. However, in the view of the board another feature of claim 1 is not disclosed in document D10:

Step b) of claim 1 reads: "(b) washing the ion exchange material with an intermediate buffer at a second conductivity which is greater than the conductivity of the loading buffer so as to elute the contaminant from the ion exchange material;".

Thus, according to claim 1 the mandatory result of washing the column with the intermediate buffer is the elution of the contaminant.

55. Document D10 does not explicitly refer to any contaminant from which the recombinant HPS is to be purified. However, it is stated at page 2552, second

column: "This type of chromatography allowed the separation of the poorly carboxylated material and yielded a purity of up to 95% in one step."

56. Human protein S is a vitamin K-dependent plasma protein with cofactor activity. The protein purified from human plasma contains approximately ten residues of gamma-carboxy-glutamate (Gla). The cofactor activity of HPS requires the Gla residues. Document D10 relates to recombinantly produced HPS. It is reported that nearly all of recombinant HPS secreted from the 293 cell line is fully carboxylated and therefore has a high cofactor activity. In contrast, HPS produced by the AV12 cell line secretes completely and incompletely gamma-carboxylated HPS. The HPS with less Gla residues had reduced cofactor activity compared to the fully carboxylated species.

57. The board considers that, in view of the teaching in document D10 that HPS function critically depends on full gamma-carboxylation and the statement that poorly carboxylated material could be separated (see above point 55), the skilled person would derive that it is the non-fully carboxylated HPS which is considered as the contaminant in document D10.

58. Document D10 discloses that the non-fully carboxylated HPS elutes after the fully carboxylated material (page 2549, right column, first full paragraph and page 2552, right column, first full paragraph, lines 11 to 22). Thus, the contaminant elutes after the desired protein HPS. Thus, step (b) of claim 1 is not disclosed in document D10. Therefore, document D10 does not disclose the subject-matter of claim 1.

Document D27

Admission of document D27

59. On 25 February 2008, i.e. during the appeal proceedings a third party filed a submission pursuant to Article 115 EPC. It contained inter alia document WO 9635718, i.e. document D27 in these proceedings, disclosing in Example 6 a process for the purification of erythropoietin with anion exchange chromatography. Moreover, the submission contained a table setting out the compositions of the buffers used in the process according to Example 6 as well as conductivity values determined for these buffers.

60. Document D27 is a late-filed document. Hence, its admissibility is at the discretion of the board pursuant to Article 114(2) EPC (see also headnote of decision T 390/07 of 20 November 2008). Considerations to be made when deciding how to exercise the discretion have been explained above in point 2.

61. The observations under Article 115 EPC were filed more than 1 1/2 years before the oral proceedings so that the parties and the board had sufficient time to consider the document. In fact, appellant I has reacted already in the written proceedings to the submission of the third party by filing amended claims and a declaration, i.e. the declaration by Dr Dowd, document D29. Also the board has studied the submissions in order to get a view of their relevance, in particular of document D27 and has notified the parties that it considers document D27 to be relevant.

None of the parties has requested that the submission of the third party, in particular document D27, be disregarded.

Thus, the board decides to admit the submission of the third party into the proceedings.

Remittal of the case to the department of first instance for consideration of document D27

62. Article 111(1) EPC gives the boards of appeal the discretion either to "exercise any power within the competence of the department which was responsible for the decision appealed" or to "remit the case to that department for further prosecution". It follows from this provision that a board is not obliged to remit a case for consideration to the first instance only because new material has been submitted which has not been considered during the first instance proceedings.

63. This is also not derivable from the Enlarged Board of Appeal's decision G 9/91 (OJ EPO, 1993, 408). It is stated in point 18 of the reasons: "The purpose of the appeal procedure inter partes is **mainly** to give the losing party the possibility of challenging the decision of the Opposition Division on its merits." It is true that in the following passage the judicial and therefore less investigative nature of the appeal proceedings is highlighted in the decision G 9/91, however only to remark a contrast with the opposition proceedings which are of an administrative and therefore more investigative nature.

64. When exercising the discretion given by Article 111(1) EPC the boards balance the public interest in procedural economy with the entitlement of the parties to fair proceedings.

For the reasons given above in point 61, the board considers it appropriate to deal with document D27 itself, thus avoiding the delay in reaching a final decision which would be entailed if the case was remitted.

65. Thus, appellant I's request for remittal to the first instance for consideration of document D27 is refused.

Substantive issues

66. According to the values in the table included in the submissions pursuant to Article 115 EPC the conductivity of the buffers used in Example 6 of document D27 is as follows (according to the terms used in the patent):

- loading buffer: 1.8 mS/cm
- intermediate buffer: 2.7 mS/cm
- wash buffer: 1.8 mS/cm
- elution buffer: 9.6 mS/cm.

In view of these values the third party considered the disclosure in Example 6 in document D27 as novelty-destroying for the subject-matter of claim 1.

67. Dr Dowd in document D29 discloses re-determined values for the conductivities of the buffers used in the process of Example 6 of document D27. He found that the

conductivities of the loading, wash and elution buffers were approximately the same as indicated in the submission by the third party. However, it turned out that the conductivity of the intermediate buffer was between 0.92-1.04 mS/cm and thus significantly lower than the value of 2.7 mS/cm disclosed in the third party's submission. Hence, on the basis of Dr Dowd's experiments document D27 would not be novelty-destroying because feature b) of claim 1 would not be fulfilled, i.e. the conductivity of the intermediate buffer would not be greater than the conductivity of the loading buffer.

68. In the third party's submission it is not stated how the buffers were prepared, how conductivity was determined and who carried out the experiments.

69. In contrast, in relation to the conductivity data in document D29, it is known from the declaration that the experiments were performed by scientists of Genentech, how buffers were made and that two series of measurement were carried out (which led to approximately the same values) and how measurements were conducted, i.e. for example, the apparatus or temperature. Thus, taking also into account that the evidence was provided in the form of a declaration at the end of which Dr Dowd declares that "all statements made in this declaration from his own knowledge are true and all statements made on information and belief are believed to be true" the board considers the evidential weight of Dr Dowd's declaration to be higher than that of the third party's submission. The board notes that Dr Dowd was also present at the oral

proceedings so that the board could have addressed him in case of doubt about the data.

70. Appellant II did not comment on the conductivity data provided by Dr Dowd, either during the written proceedings or at the oral proceedings. Thus, in fact, the values are uncontested.

71. Under these circumstances the board considers the conductivities given in the declaration of Dr Dowd as correct. Thus, the disclosure in Example 6 of document D27 does not match the conductivity pattern according to claim 1 (see point 67 above).

72. The board thus concludes that the subject-matter of claim 1 and claims dependent on it is not anticipated by the disclosures in any of documents D7, D10 or D27.

73. The subject-matter of independent claim 2 and claims dependent on it is novel over the disclosure in documents D7, D10 and D27 because none of them discloses a method for purifying an antibody. The subject-matter of independent claim 22 is novel in view of documents D7, D10 and D27 because none of them discloses the claimed method steps in combination with cation chromatography. Appellant II did not raise objections with regard to claims 2 and 22.

74. The requirements of Article 54 EPC are fulfilled.

Inventive step

Closest prior art

75. It has been repeatedly pointed out in the case law of the boards of appeal that the closest prior art document is a document disclosing subject-matter conceived for the same purpose or aiming at the same objective as the claimed invention.

However, it has also been emphasized that "careful consideration must be given to the question whether, in the case concerned, the skilled person, taking into account all the available information on the technical context of the claimed invention would have had good reason to take this prior art as the starting point for further development (Case Law of the Boards of Appeal of the EPO, 5th edition, I.D.3.1). Furthermore it is pointed out in the case law that the aim of determining the closest prior art is to select a document allowing the assessment process to start from a situation as close as possible in reality to that encountered by the inventor (Case Law of the Boards of Appeal of the EPO, 5th edition, I.D.3.2, first paragraph) or in other words "as closest prior art a "bridgehead" position should be selected which said skilled person would realistically have taken under the circumstances of the claimed invention." (Case Law of the Boards of Appeal of the EPO, 5th edition, I.D.3.4, last paragraph). It is further noted that theses considerations should be given more weight than the commonality of technical features.

76. During the written proceedings appellant II raised objections of lack of inventive step on the basis of documents D1, D6, D7 and D10. At the oral proceedings the objections were restricted to documents D1 and D6.

77. In paragraph [0005] of the patent after explaining the general principle of ion chromatography it is emphasized that "in the past, these changes have been progressive, i.e., the pH or conductivity is increased or decreased in a single direction."

78. Moreover, it is known that generally, the desired protein is eluted after the elution of the contaminant, i.e. at a higher conductivity. This is also the situation referred to in the claims.

79. According to documents D1, D6 and D10 the elution of the **contaminant** is achieved by a change in the **charge** of the protein whereas a **conformational** change of the protein due to the binding of calcium ions is responsible for eluting the **desired protein (i.e.** pseudoaffinity chromatography mode, see also point 51 above). Normally, with this "mixed" way of chromatography, the increase of the salt concentration in the buffers is not progressive. In particular, the conductivity of the wash buffer is lower than that of the intermediate buffer.

80. Seeing the circumstances of the present invention as disclosed in paragraph [0005] of the patent, i.e. that the starting point for the invention is considered to be the "standard" ion chromatography as opposed to ion chromatography conducted in a pseudo-affinity mode, the board comes to the conclusion that the skilled person

would not have realistically selected one of documents D1, D6 or D10 as closest prior art documents.

81. As to document D7, it discloses the desorption of the contaminant, endotoxin A, from the ion chromatography column by virtue of the activity of a non-ionic detergent. Therefore, document D7 also does not relate to "classical" ion chromatography referred to in the patent and consequently does not represent the closest prior art document.

82. In the board's view, the skilled person would consider the prior art referred to in paragraph [0005] of the patent in suit as the most promising springboard towards the invention, namely ion exchange chromatography where conductivity is changed in a linear increasing manner. Such an ion chromatography process is, for example, disclosed in document D3. Ascites fluid containing monoclonal antibody BCD-1 is loaded onto an ion exchange column and the antibody and impurities separated by elution with a linear gradient from 0 to 0.5 M NaCl (pages 152 and 153).

Problem and solution

83. The problem to be solved is to improve this well-known chromatography process such that the purity of the desired protein is increased by achieving a better separation of contaminant and protein.

84. The solution to this problem as claimed consists in a process where conductivity is not progressively increased for elution, but where after elution of the contaminant, the conductivity of the buffer is

decreased, before increasing it again for elution of the desired protein.

85. The patent provides one example, i.e. the separation of deamidated variants of anti-HER2 antibody from the "wild-type" anti-HER2 antibody. By using the claimed method the content of the contaminant could be reduced to about 13% or less when compared to 25% when the standard method was used (paragraph [0107]. It is stated in paragraph [0108]: "It was discovered that by going back to lower conductivity as used initially the elution of the deamidated anti-HER2 antibody continued, without significant anti-HER2 antibody-product elution".

86. The board considers that this example is sufficient to demonstrate that the claimed solution, which the board considers to be a conceptually new way of conducting "standard" ion chromatography, has actually been achieved by the patent.

Obviousness

87. Appellant II argues that the subject-matter of claim 1 is obvious in view of either of documents D1, D6, D7 or D10.

Thus, the question is whether or not the skilled person wanting to improve of classical way of conducting ion chromatography such that better separation of contaminant and protein is achieved would get any motivation from the disclosure in documents D1, D6 D7 or D10 to modify the "classical" buffer pattern, i.e. a progressive increase in the conductivity, such that the conductivity of the buffer corresponding to the "wash

buffer" in claim 1 is less than the conductivity of the buffer corresponding to the "intermediate buffer" in claim 1 in order to achieve better separation.

88. The prior art has to be assessed from the point of view of the skilled person on the priority date (Case Law of the Boards of Appeal of the EPO, 5th edition 2006, I.D.3.1, last sentence)

89. Each of documents D1, D6 and D10 describe processes where the conductivity of the buffer corresponding to the wash buffer in claim 1 is less than the conductivity of the buffer corresponding to the intermediate buffer.

Document D1

90. However, document D1 as a whole teaches that the improvement in the purity of factor IX after purification with anion chromatography is achieved by conducting the elution of factor IX in a pseudo-affinity mode because this achieves rather specific elution of active factor IX (page 8, lines 18 to 21 and page 11, lines 10 to 12).

On page 11, line 20 to 21 of document D1 the method is disclosed in more detail. With regard to the step corresponding to the wash step in the claimed method it is stated: "The column is washed with 50mM TRIS, 100mM NaCl, pH 8.0 to **lower the conductivity in preparation for elution**." (emphasis added).

Document D6

91. Document D6 discloses that the purity of factor FVII after purification by anion chromatography is enhanced by the presence of zinc ions in at least one of the purification steps, because this **avoids the generation** of contaminants during the purification steps. Thus, it is for example stated on page 3, lines 1 to 5 and 7 to 9:

"It is therefore the purpose of the present invention to provide a purification process for FVII by which activation and degradation is avoided or kept at an acceptable low degree with the purpose of providing a homogeneous product of high purity... . [...] It has now surprisingly been found that addition of zinc ions can be used to control the autoactivation of FVII and to impede the degradation of FVII/FVIIA during purification by means of chromatographic column materials."

Document 10

92. Document D10 reports in the discussion-section that it has been difficult to identify mammalian cell lines expressing functionally active and correctly modified vitamin K-dependent proteins due to the number and complexity of post-translational modifications. The document discloses the expression of cDNA for human protein S, which is a vitamin K-dependent protein, in two mammalian cell lines and the characterization of the HPS isoforms secreted from the two cell lines. The document reports that fully functional HPS could be separated from a less functional isoform by running the

ion chromatography column in a pseudoaffinity mode for elution of the active form (see also above point 51). Hence, as with document D1, the improvement transpiring from document D10 is the fact that pseudo-affinity mode is used instead of the classical charge-induced elution.

Document D7

The objective of document D7 is to provide a highly purified protein A which is especially free of endotoxin contamination (page 3, lines 14 to 15). The removal of endotoxin is achieved by the inclusion of a non-ionic detergent in the washing buffer. It is stated on page 4, lines 17 to 20: "The use of non-ionic detergent allows the disassociation of protein A and endotoxin resulting in a reduction of less than 1.0 E.U./mg protein A, as opposed to the >10 E.U./mg protein A levels which are obtained when no detergent is used in the process." Thus, it is taught in document D7 that the contaminant can advantageously be removed by inclusion of a detergent, but not by modifying buffer conductivities.

93. In summary, the board notes firstly, that only one of the documents gives a reason as to why the conductivity of the buffer is decreased before elution, i.e. document D1 states that it is for preparation of the elution. However, this statement does not suggest to the skilled person that this step would be good for achieving better separation of protein and contaminant. Secondly, the documents teach that an increase in the purity of a desired protein can be achieved by using pseudo-affinity chromatography or by including zinc ions or a non-ionic detergent in the buffer. Thus, when

considering the whole disclosure of each of the documents and at the priority date of the patent, i.e. without the knowledge of the invention, the skilled person would not be motivated by any of them, alone or in combination, to modify the classical ion chromatography procedure in such a way as to arrive at the method now claimed. Consequently, the subject-matter of claim 1 is not obvious in the light of any of documents D1, D6, D7 and D10.

94. This conclusion also applies to independent claims 2 and 22 insofar as they relate to the buffer pattern as recited in claim 1. These claims encompass as a further embodiment, the purification by a change of pH. No arguments have been presented with regard to this aspect of the claims.

95. It has been established by case law such as represented by decision T 939/92 ("Agrevo"; OJ EPO 1996, 309) that everything falling within a valid claim has to be inventive, i.e. the technical effect on which acknowledgment of inventive step relies must be produced by essentially all embodiments of the claim.

The appellant relies on this decision and argues that this was not so with regard to claim 1 since even the patent teaches (Figure 5 and page 16, lines 39 to 42) that only some types of contaminants can be removed by the method, whereas others are unaffected.

96. The claim at issue in the case underlying decision T 939/92 was directed to a class of compounds. The effect that they are stated to have and on which the inventive contribution relied, i.e. herbicidal activity,

was derivable from the description, but not a feature of the claims. Thus, the compounds were not functionally restricted. In the present case, the claims relate to a method for purification. The board interprets such a claim to mean that any of its embodiments is in fact suited to achieve that stated purpose, i.e. they are suited to achieve purification. Thus, the question of whether certain embodiments of the claim do or do not achieve the expected technical effect does not arise. The board considers therefore, that the considerations of decision T 939/92 in the assessment of inventive step do not apply here in the context of inventive step. Rather in the present case they arise in the context of the evaluation of Article 83 EPC when considering the question of whether or not the claimed subject-matter can be carried out over the whole scope without undue burden, they therefore have been dealt with there (see points 25 to 31 above).

97. In conclusion the subject-matter of claims independent claims 1, 2 and 22 and claims dependent thereon involves an inventive step. The requirements of Article 56 EPC are fulfilled.

Order

For these reasons it is decided that:

1. The decision under appeal is set aside.

2. The case is remitted to the department of first instance with the order to maintain the patent with the following claims and a description to be adapted thereto:

   Claims 1-22 of the Main Request filed at the oral proceedings on 23 September 2009.

The Registrar: The Chair:

P. Cremona U. Kinkeldey