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(71) Applicant: **KABUSHIKI KAISHA KOBE SEIKO
SHO**
**3-18, Wakinohamacho 1-chome Chuo-ku
Kobe-shi Hyogo-ken 651(JP)**

(72) Inventor: **NISHIDA, Tomoaki**
1588-67, Sakuragaoka Fujishiro-cho
Kitasoma-gun Ibaragi-ken 300-15(JP)
Inventor: **KASHINO, Yoshinori**

**12-18, Hakomatsu 3-chome Higashi-ku
Fukuoka-shi Fukuoka-ken 812(JP)**

Inventor: **TAKAHARA, Yoshimasa**
**5-601, 18-ban, Kasuga 2-chome
Tsukuba-shi Ibaragi-ken 305(JP)**

Inventor: **SAKAI, Kokki**
**3693-33, Fukuma-cho
Munakata-gun Fukuoka-ken 811-32(JP)**

Inventor: **KONDO, Ryuichiro**
**8-408, 23-ban, Kashii 1-chome Higashi-ku
Fukuoka-shi Fukuoka-ken 813(JP)**

(74) Representative: **Lewin, John Harvey et al**
Elkington and Fife Prospect House 8
Pembroke Road
Sevenoaks, Kent TN13 1XR(GB)

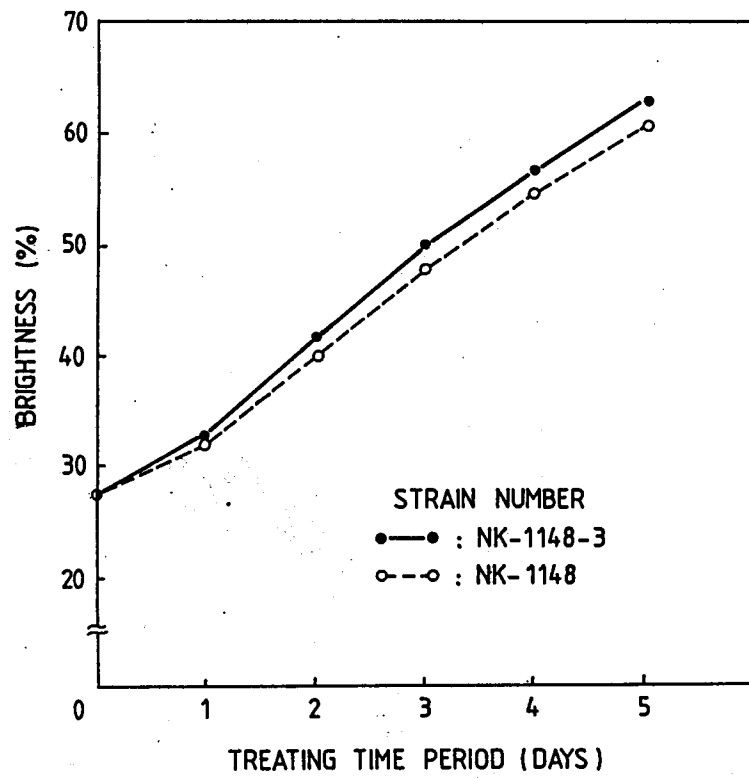
(54) **PROCESS FOR PRODUCING PULP.**

(57) A process for producing pulp substantially without the necessity for adding a nutrient or a cellulose degradation inhibitor, which comprises treating the raw material in the pulping step with a microorganism which grows well in a medium containing lignin as a sole carbon source. Since the addition of a nutrient or a cellulose degradation inhibitor is indispensable in the conventional microbial treatment, the present invention which can dispense with the addition of these substances can achieve a remarkable cost reduction as well as an energy saving by virtue of the microbial treatment.

EP 0 464 221 A1

FIG. 1

RELATIONSHIP BETWEEN TREATING
TIME PERIOD AND BRIGHTNESS



Field of the Invention

This invention relates to methods for producing pulp, comprising microbial treatment, in either or both of pulping process and bleaching process, using a microorganism which grows well in a culture medium containing lignin as a single carbon source.

The present invention relates to methods for producing pulp, comprising microbial treatment substantially without adding nutrients or inhibitors of cellulose degradation at any one stage of the process of producing pulp.

The present invention enables to produce pulp in good quality, by microbial treatment using a microorganism which grows well in a culture medium containing lignin as a single carbon source at a process for producing pulp, thereby suppressing cellulose degradation to the minimum.

The present invention enables pulping or bleaching, substantially without adding nutrients or inhibitors of cellulose degradation, for example glucose, at a process of producing pulp, by using a microorganism with excellent lignin-degrading activity and a high selectivity, whereby the present invention can provide economical and industrial methods for producing pulp in remarkably energy-saving manner.

Prior Art and Problems

Since considerably early days, a great number of research efforts have been carried out on pulping or bleaching for the process of producing paper and pulp by using microorganisms.

The Japanese Patent Laid-open No. 46903/1975 proposes a method for producing cellulose pulp, comprising degradation of lignin under the condition to substantially degrade lignin, by using a microorganism having a production potential of a lignin-degrading enzyme.

However, the method has never been put to industrial use, because the degree of lignin degradation is so low due to the extremely low lignin-degrading activity of the microorganism used, and because the addition of sugars and nitrogen compounds is required added due to the suppression of cellulose assimilation by the microorganism.

The bleaching of pulp with *Phanerochaete chrysosporium* has been also reported (Biotechnol Lett., 1, 347-353(1979)), but it has neither been practiced industrially because of the low lignin-degrading activity and the use of the large amount of an inhibitor of cellulose degradation.

Means to Solve the Problems

The present inventors have investigated intensively in order to develop a method for pulping or bleaching with microbial treatment, without causing cellulose degradation and adding a nutrient and inhibitor of cellulose degradation. Consequently, they have achieved the object in accordance with the present invention.

That is, in accordance with the present invention, it is the use of a microorganism which grows well in a culture medium containing lignin as a single carbon source that realizes pulping and bleaching of wood chips, pulp after refining and unbleached pulp, substantially without adding nutrients or inhibitors of cellulose degradation, in the economical and tremendously energy-saving manner.

Brief Description of the Drawings

Fig. 1 shows the increase in brightness (%) after the microbial treatment for 1 to 5 days in Example 5.

The microorganism to be used in the present invention is from a microbial strain, well grown by inoculation and culture in a culture medium containing lignin as a single carbon source.

As the culture medium, there may be prepared an agar medium to which is added as a single carbon source, about 1 to 10% of lignin, preferably 2 to 4% of lignin.

An isolating source collected from a natural source is dispersed at an appropriate concentration in the culture medium, and cultured at 25 to 35 °C, to collect a colony exhibiting a good growth, which is to be an effective microorganism to be used in the present invention.

The present inventors have previously isolated the strains NK-1148 (FERM BP-1859) and NK-729W (FERM BP-1860), which are among the microorganisms very effective for the present invention.

The mycological characteristics of NK-1148 strain (FERM BP-1859) are shown as follows.

(1) State of Growth in Culture Media

Type of Medium	State of Growth
Malt extract agar medium	+++
Potato-glucose agar medium	+++
Czapek's agar medium	+
Sabouraud's agar medium	++
Synthesized mucor agar medium	++
YpSs agar medium	+++
Glucose-dried yeast agar medium	+++

Note-1 pH of the medium: 5.0 (before sterilization in autoclave)

Note-2 Culture conditions: 28 °C x 7 days

Note-3 State of growth

weak : +
medium : ++
abundant : +++

(2) Physiological and Morphological Properties

① pH range for the growth (Cultivation in a potato-glucose agar medium at 28 °C for 4 days)

Grows at pH near 3 - 9, but never grows at pH 2 or pH 10. The optimum pH is near 4 to 6.

② Temperature range for the growth (Cultivation in a potato-glucose agar medium at pH 5 for 4 days)

Grows at temperatures near 10 - 45 °C, but never grows at 50 °C. The optimum temperature range is near 28 - 37 °C.

③ Phenol oxidase reaction (Cultivation at 28 °C for 4 days)

Shows weak or negative response.

④ Morphology of colony (Cultivation in a potato-glucose agar medium at pH 5 at 28 °C for 4 days)

White and felt-like.

The mycological characteristics of NK-729W strain (FERM BP-1860) are shown as follows.

(1) State of Growth in Culture Media

Type of Medium	State of Growth
Malt extract agar medium	+++
Potato-glucose agar medium	+++
Czapek's agar medium	+
Sabouraud's agar medium	++
Synthesized mucor agar medium	++
YpSs agar medium	+++
Glucose-dried yeast agar medium	+++

Note-1 pH of the medium: 5.0 (before sterilization in autoclave)

Note-2 Culture conditions: 28 °C x 7 days

Note-3 State of growth

weak : +
medium : ++
abundant : +++

(2) Physiological and Morphological properties

① pH range for the growth (Cultivation in a potato-glucose agar medium at 28 °C for 4 days)

Grows at pH near 3 - 7, but never grows at pH 2 or pH 8. The optimum pH is near 4 to 5.

② Temperature range for the growth (Cultivation in a potato-glucose agar medium at pH 5 for 4 days)

Grows at temperatures near 10 - 32 °C, but never grows at 37 °C. The optimum temperature range is near 20 - 30 °C.

③ Phenol oxidase reaction (Cultivation at 28 °C for 4 days)

Shows positive response.

④ Morphology of colony (Cultivation in a potato-glucose agar medium at pH 5 at 28 °C for 4 days)

White and hairy.

⑤ Morphology of fruit body

Size : 2 - 5 mm diameter

Shape : Inverted cup shape (nose shape)

Edge or surface : Edge turned inwardly, surface color of yellow black, having brown fleece or hair over the entire surface

Surface of tubular pore : Pale white gray, recessed in an upturned dish shape, with small pore

Texture : Soft leather-like texture, substantially white

⑥ Spore shape

About 3 - 4 x 1 μm, sausage-like shape, colorless and smooth.

The microorganism to be used in the present invention may be NK-1148 strain or NK-729W strain

isolated by the present inventors, but preferably there may be used a selectively isolated strain growing well using lignin as a single carbon source, obtained with or without mutation of these two strains, or a strain growing well using lignin as a single carbon source and having been isolated from nature.

In accordance with the present invention, the microorganism growing well using lignin as a single carbon source, represents, for example, a microorganism capable of bleaching an unbleached kraft pulp up to a brightness of 45 % or more, preferably 50 % or more, more preferably 60 % or more, with no reduction in the strength of the pulp.

The microorganism to be used in the present invention can be cultured in any one of a culture medium containing lignin as a single carbon source, a general culture medium for basidiomycetes and fungi without containing lignin, and a culture medium containing wood powder, wood chips and pulp.

The type of pulp is generally classified in the following three.

- ① Mechanical pulp fiberized by mechanical treatment of wood.
- ② Semicheical pulp obtained by chemical and mechanical treatments in combination.
- ③ Chemical pulp with most of lignin removed through chemical treatment.

The present invention is to produce individual pulp corresponding mechanical pulp (the degree of lignin degradation below 35 %), semicheical pulp (the degree of lignin degradation of not less than 35% to less than 75 %), and chemical pulp (the degree of lignin degradation of not less than 75%), by replacing a part or the entire part of the chemical treatment or mechanical treatment in the processes of producing pulp i.e.,

① to ③, with the microbial treatment of the present invention.

That is, the following processes are fundamentally included in the present invention.

A: Wood chips are directly treated with the microorganism of the present invention, to degrade the lignin component in the wood chips to produce unbleached pulp.

B: Pulp from the wood chips refined at a light degree is treated with the microorganism of the present invention, to degrade the lignin component in the pulp to produce unbleached pulp.

C: The wood chips after chemical treatment at a light degree are treated with the microorganism of the present invention, to degrade the lignin component in the pulp described above to produce unbleached pulp.

D: The pulp obtained by lightly refining wood chips is chemically treated lightly, and then are treated with the microorganism of the present invention, to degrade the lignin component in the pulp to produce unbleached pulp.

E: The pulp obtained by chemically treating wood chips lightly and refining the wood chips lightly is treated with the microorganism of the present invention, to degrade the lignin component in the pulp to produce unbleached pulp.

F: The unbleached pulp obtained in any one of A to E is further treated by a light chemical treatment and/or light refining to produce unbleached pulp.

In the microbial treatment in the processes A to F, there may be used the microorganism growing well in the culture medium containing lignin as a single carbon source. The microorganism can selectively degrade the lignin component in wood and uses the wood lignin as a nutrient. It is therefore possible to carry out the aforementioned microbial treatment without adding an inhibitor of cellulose degradation.

The degree of the chemical treatment to a light degree and the refining treatment to a light degree in the processes A to F is appropriately determined by a predetermined lignin content, depending on the type of the unbleached pulp including mechanical pulp, semicheical pulp and chemical pulp, or in any type of the pulp.

The microorganism to be used in the present invention has a far greater lignin-degrading activity than the lignin-degrading microorganisms conventionally known. Hence, the present invention enables the substitution of all stages of the chemical treatment and the refining in the conventional processes of producing mechanical pulp, semicheical pulp and chemical pulp, with the microbial treatment, along with the marked decrease in the degree of the chemical treatment and the refining. In other words, the process of producing pulp in accordance with the present invention can decrease the amount of chemicals, and is appropriate for production of high-quality pulp in energy-saving manner.

As the fundamental processes in the process of producing pulp according to the present invention, there have been described herein A to F. But they are just representative, so it is possible to appropriately combine the microbial treatment with the microorganism of the present invention, with other treatments.

Intensely colored lignin generally remains in unbleached chemical pulp and unbleached semicheical pulp. In case of using these unbleached pulp for papers for the use requiring a higher brightness, therefore, the pulp is transferred to the bleaching process to remove the remaining lignin, to increase the brightness.

In such case, in accordance with the present invention, unbleached pulp is treated with the microorganism growing well in a culture medium containing lignin as a single carbon source, to degrade and remove

the remaining lignin in the unbleached pulp, for the bleaching of the unbleached pulp.

The unbleached pulp may be any one of the unbleached chemical pulp and unbleached semichemical pulp by conventional methods, and the unbleached pulp corresponding to chemical pulp and those corresponding to semichemical pulp, produced with the microbial treatment of the present invention. The bleaching of the present invention may be applied to the unbleached mechanical pulp by the conventional methods, and the unbleached pulp corresponding to mechanical pulp produced through the microbial treatment of the present invention. However, since a great amount of chlorine-based bleaching agents is used for bleaching of unbleached chemical and semichemical pulp, the present bleaching is effectively applied to the unbleached pulp corresponding to unbleached chemical and semichemical pulp, from the standpoint of pollution control.

In accordance with the present invention, the bleaching process may be carried out entirely as the bleaching with the use of the microorganism, but a combination of the present bleaching with other bleaching methods may be also possible. The bleaching of the present invention can achieve a high standard of safety due to its microbial treatment.

Pulping and/or bleaching can be carried out through by adding the cultured microorganism to about 1/10000 to 10/100 of wood chips or pulp, and culturing the mixture at about 20 to 35 °C for 3 to 90 days, without adding any nutrient or inhibitor of cellulose degradation to wood chips or various pulp. Optional addition of a small amount of inhibitors of cellulose degradation may be done.

Example 1

The culture medium containing 1.0 g of beech wood powder (60 - 80 mesh) and 2.5 ml of water, placed in a 50 ml flask, is heated and sterilized at 120 °C for 15 minutes, into which is inoculated NK-1148 strain (FERM BP-1859) and cultured at 28 °C for a week. The resulting mycelia are suspended in water.

Alternatively, the culture medium containing 2.0 % of milled wood lignin from white birch, 0.2 % of $\text{NH}_4\text{H}_2\text{PO}_4$ and 1.6 % of agar is heated and sterilized at 120 °C for 15 minutes, which is then aseptically divided by 20 ml each into petri dishes (90 ml in diameter).

The mycelia in suspension were added to the culture medium and cultured at 28 °C for two weeks, to isolate a strain exhibiting good growth, which was defined an isolated strain A. The isolated strain A was designated as NK-1148-3 strain and has been deposited in Fermentation Research Institute, Agency of Industrial Science and Technology, under the accession number of FERM BP-3220.

Example 2

The isolated strain NK-1148-3 obtained in Example 1 was inoculated into the culture medium containing 1.2 % of potato-dextrose broth commercially available (DIFCO Co., Ltd.) after sterilization at 120 °C for 15 minutes, and cultured at 28 °C for a week, which was used as a seed culture.

Example 3

In the process of producing mechanical pulp, 10 kg of pulp obtained through a first refining of beech wood chips at a light degree was mixed with 24.5 l of water, and sterilized at 120 °C for 15 minutes, to which was added 0.5 kg of the seed culture obtained in Example 2 and cultured under aeration at 28 °C for a week. Subsequently after a second refining, a mechanical pulp with a high strength could be obtained in an extremely energy-saving manner. The properties of the obtained biomechanical pulp are shown in the following Table 1.

Table 1 Properties of biomechanical pulp

	Refining energy (KWH/pulp (t))	Quality of bleached pulp		
		kPa·m ² /g Burst index	N·m/g Tensile index	mN·m ² /g Tear index
Mechanical pulp	2000	0.52	9.6	1.21
Biomechanical pulp	710	1.15	20.3	2.53

Example 4

NK-1148 strain and the isolated strain NK-1148-3, obtained in Example 1, were separately inoculated into each culture medium mixed with 10 kg of an unbleached pulp (eucalyptus) and 25 l of water, which medium had been treated and sterilized in advance at 120 °C for 15 minutes, and cultured under aeration at 28 °C for two weeks. The resulting cultures were individually used as a seed culture.

Example 5

For bleaching of chemical pulp, 10 kg of a unbleached kraft pulp (eucalyptus) was mixed with 25 l of water and sterilized at 120 °C for 15 minutes, to which were separately added 1 kg of each of the seed cultures obtained in Example 4, together with 0.5 kg of glucose, and mixed for culture under, and cultured under aeration at 28 °C for 1 to 5 days, leading to the production of a bleached kraft pulp. The enhancement of the brightness during the microbial treatment period for 1 to 5 days is shown in Fig. 1.

Claims

1. A method for producing pulp from wood by using a microorganism, comprising microbial treatment, in either pulping process or bleaching process or both, using the microorganism which grows well in a culture medium containing lignin as a single carbon source.
2. A method for producing unbleached pulp, comprising degrading at least partially lignin present in wood chips by using a microorganism which grows well in a culture medium containing lignin as a single carbon source.
3. A method for producing unbleached pulp, comprising refining wood chips to a light degree to produce pulp and degrading at least partially the lignin present in the obtained pulp by using a microorganism which grows well in a culture medium containing lignin as a single carbon source.
4. A method for producing unbleached pulp, comprising treating wood chips with a chemical to a light degree and degrading at least partially the lignin present in the resulting wood chips by using a microorganism which grows well in a culture medium containing lignin as a single carbon source.
5. A method for producing unbleached pulp, comprising refining wood chips to a light degree to produce

pulp, treating the obtained pulp with a chemical to a light degree, and degrading at least partially the lignin present in the resulting pulp by using a microorganism which grows well in a culture medium containing lignin as a single carbon source.

- 5 **6.** A method for producing unbleached pulp, comprising treating wood chips with a chemical to a light degree, refining the resulting wood chips to produce pulp, and degrading at least partially the lignin present in the obtained pulp by using a microorganism which grows well in a culture medium containing lignin as a single carbon source.
- 10 **7.** The method for producing pulp according to claims 2 to 6, wherein the unbleached pulp is further treated with a chemical and/or refined after the microbial treatment.
- 8.** The method for producing pulp according to claims 1 to 7, wherein the microbial treatment is carried out in the state where a nutrient or inhibitor of cellulose degradation is not substantially added.
- 15 **9.** A method for producing bleached pulp, comprising carrying out at least a part of bleaching treatment of unbleached pulp in bleaching process, using a microorganism which grows well in a culture medium containing lignin as a single carbon source.
- 20 **10.** The method for producing bleached pulp according to claim 9, wherein the bleaching treatment using the microorganism is carried out in the state where a nutrient or inhibitor of cellulose degradation is not substantially added.

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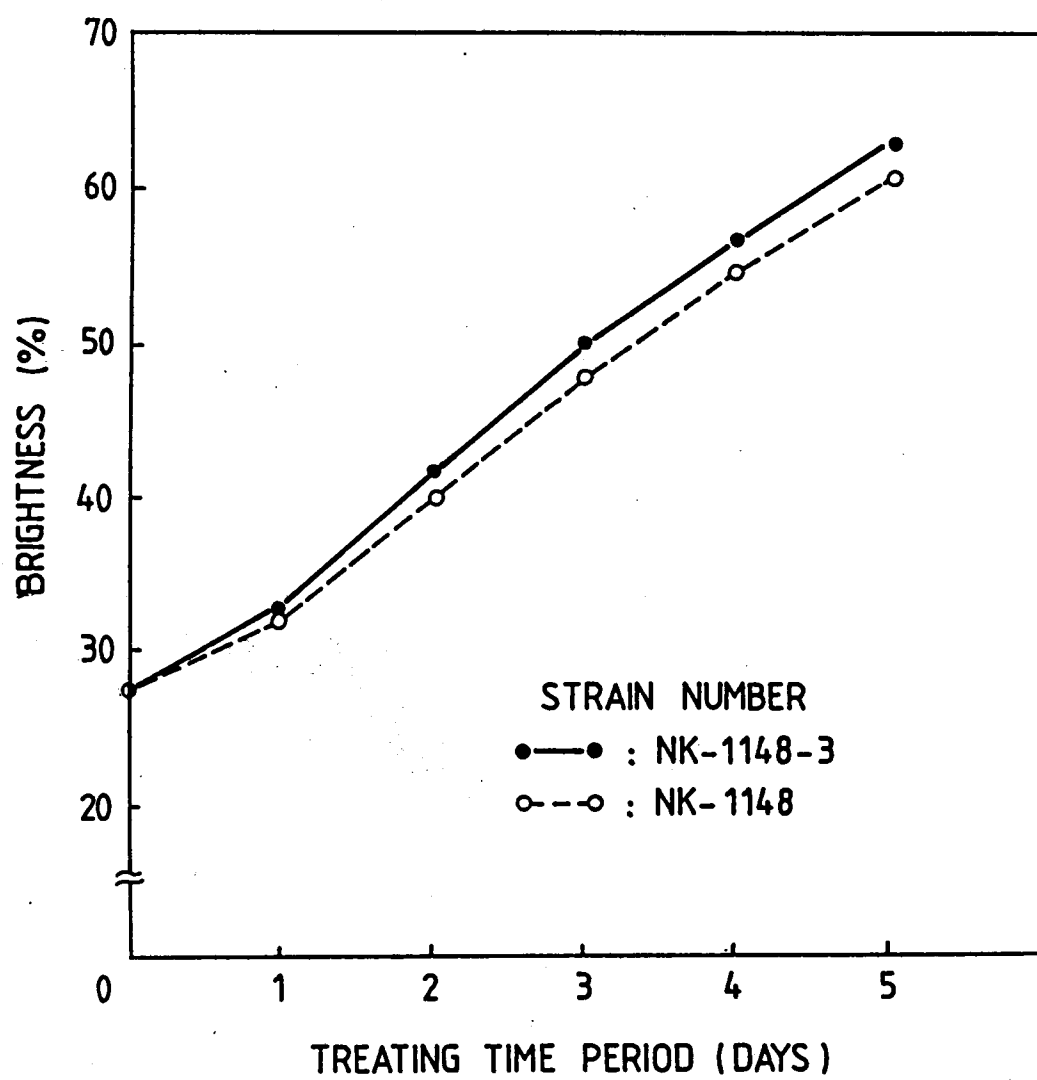
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*FIG. 1*RELATIONSHIP BETWEEN TREATING
TIME PERIOD AND BRIGHTNESS

INTERNATIONAL SEARCH REPORT

International Application No PCT/JP91/00048

I. CLASSIFICATION OF SUBJECT MATTER (if several classification symbols apply, indicate all) ⁶		
According to International Patent Classification (IPC) or to both National Classification and IPC		
Int. Cl ⁵ D21C3/22, 3/00, C12S3/08, D21B1/14, D21C9/10, D21D1/20, C12N1/22		
II. FIELDS SEARCHED		
Minimum Documentation Searched ⁷		
Classification System	Classification Symbols	
IPC	D21C3/22, 3/00, 5/00, C12S3/08, C12N1/22, D21B1/14, D21C9/10, D21D1/20	
Documentation Searched other than Minimum Documentation to the Extent that such Documents are Included in the Fields Searched ⁸		
III. DOCUMENTS CONSIDERED TO BE RELEVANT ⁹		
Category ¹⁰	Citation of Document, ¹¹ with indication, where appropriate, of the relevant passages ¹²	Relevant to Claim No. ¹³
Y	JP, A, 62-500214 (Svenska Träforsknings- stitutet), January 29, 1987 (29. 01. 87) & WO, A1, 86/1843 & AU, A1, 4959285 & NO, A, 861809 & DK, A, 231086 & FI, A, 871177 & EP, A1, 245237 & SE, B, 460363	1-10
Y	JP, A, 63-500632 (Repligen Corp.), March 10, 1988 (10. 03. 88) & PT, A, 82979 & WO, A1, 87/0550 & AU, A1, 6138586 & FI, A, 871088 & NO, A, 871060 & DK, A, 129587 & EP, A1, 229171 & US, A, 4687741 & BR, A, 8606809 & PH, A, 22286	1-10
Y	JP, A, 63-500604 (Repligen Corp.), March 3, 1988 (03. 03. 88) & PT, A, 82980 & WO, A1, 87/0564 & AU, A1, 6141286 & FI, A, 871089 & NO, A, 871061 & EP, A1, 231275 & US, A, 4687745 & BR, A, 8606775 & CA, A1, 1249783 & YU, A, 125386	1-10
¹⁰ Special categories of cited documents: "A" document defining the general state of the art which is not considered to be of particular relevance "E" earlier document but published on or after the international filing date "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified) "O" document referring to an oral disclosure, use, exhibition or other means "P" document published prior to the international filing date but later than the priority date claimed "T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention "X" document of particular relevance: the claimed invention cannot be considered novel or cannot be considered to involve an inventive step "Y" document of particular relevance: the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art "&" document member of the same patent family		
IV. CERTIFICATION		
Date of the Actual Completion of the International Search	Date of Mailing of this International Search Report	
March 18, 1991 (18. 03. 91)	April 8, 1991 (08. 04. 91)	
International Searching Authority	Signature of Authorized Officer	
Japanese Patent Office		

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Y	US, A, 235662 (Int. Paper Co.), June 8, 1988 (08. 06. 88)	1-10

V. ☐ OBSERVATIONS WHERE CERTAIN CLAIMS WERE FOUND UNSEARCHABLE ¹

This international search report has not been established in respect of certain claims under Article 17(2) (a) for the following reasons:

1. ☐ Claim numbers , because they relate to subject matter not required to be searched by this Authority, namely:
2. ☐ Claim numbers , because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:
3. ☐ Claim numbers , because they are dependent claims and are not drafted in accordance with the second and third sentences of PCT Rule 6.4(a).

VI. ☐ OBSERVATIONS WHERE UNITY OF INVENTION IS LACKING ²

This International Searching Authority found multiple inventions in this international application as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims of the international application.
2. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims of the international application for which fees were paid, specifically claims:
3. ☐ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claim numbers:
4. ☐ As all searchable claims could be searched without effort justifying an additional fee, the International Searching Authority did not invite payment of any additional fee.

Remark on Protest

- ☐ The additional search fees were accompanied by applicant's protest.
- ☐ No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM THE SECOND SHEET

	& EP, A, 345715 & PT, A, 90776 & AU, A, 8936045 & NO, A, 8902335 DK, A, 8902784 & BR, A, 8902665 & FI, A, 8902803 & JP, A, 02-80687 ZA, A, 8904239	
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FURTHER INFORMATION CONTINUED FROM THE SECOND SHEET

Y	JP, A, 53-79086 (General Electric Co.), July 13, 1978 (13. 07. 78) & US, A, 734325 & DE, A, 2746872 & NL, A, 7711451 & SE, A, 7711780 & FR, A, 2368536 & GB, A, 1560021 & IT, B, 1087958	1-10
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2. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims of the international application for which fees were paid, specifically claims:
3. ☐ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claim numbers:
4. ☐ As all searchable claims could be searched without effort justifying an additional fee, the International Searching Authority did not invite payment of any additional fee.

Remark on Protest

- ☐ The additional search fees were accompanied by applicant's protest.
- ☐ No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM THE SECOND SHEET

A	JP, A, 63-91077 (Oji Paper Co., Ltd.), April 21, 1988 (21. 04. 88), (Family: none)	1-10
A	JP, A, 63-249789 (Kogyo Gijutsuin-cho, Hitachi Zosen Corp.), October 17, 1988 (17. 10. 88), (Family: none)	1-10
A	JP, A, 58-180692 (Katsuaki Takahashi, Manabu Kurita, Keiya Yonemitsu), October 22, 1983 (22. 10. 83), (Family: none)	1-10

V. ☐ OBSERVATIONS WHERE CERTAIN CLAIMS WERE FOUND UNSEARCHABLE ¹

This international search report has not been established in respect of certain claims under Article 17(2) (a) for the following reasons:

1. ☐ Claim numbers _____, because they relate to subject matter not required to be searched by this Authority, namely:
2. ☐ Claim numbers _____, because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:
3. ☐ Claim numbers _____, because they are dependent claims and are not drafted in accordance with the second and third sentences of PCT Rule 6.4(a).

VI. ☐ OBSERVATIONS WHERE UNITY OF INVENTION IS LACKING ²

This International Searching Authority found multiple inventions in this international application as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims of the international application.
2. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims of the international application for which fees were paid, specifically claims:
3. ☐ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claim numbers:
4. ☐ As all searchable claims could be searched without effort justifying an additional fee, the International Searching Authority did not invite payment of any additional fee.

Remark on Protest

- ☐ The additional search fees were accompanied by applicant's protest.
- ☐ No protest accompanied the payment of additional search fees.