(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
3 October 2002 (03.10.2002)

PCT

(10) International Publication Number
WO 02/077034 A3

(51) International Patent Classification[7]: C07K 14/535, A61K 38/18, C12N 15/27, C07K 7/06, 7/08

(21) International Application Number: PCT/EP02/01171

(22) International Filing Date: 5 February 2002 (05.02.2002)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:

01102617.6	6 February 2001 (06.02.2001)	EP
01103954.2	19 February 2001 (19.02.2001)	EP

(71) Applicant *(for all designated States except US)*: **MERK PATENT GMBH** [DE/DE]; Frankfurter Strasse 250, 64293 Darmstadt (DE).

(72) Inventors; and

(75) Inventors/Applicants *(for US only)*: **CARR, Francis, J.** [GB/GB]; Birchlea, The Holdings, Balmedie, Aberdeenshire AB23 8XU (GB). **CARTER, Graham** [GB/GB]; Longhills Cottage, By Newmachar, Aberdeenshire AB21 7XB (GB). **JONES, Tim** [GB/GB]; 27 Brick Row, Babraham, Cambridge CB2 4AJ (GB). **WILLIAMS, Stephen** [GB/GB]; Drumgowan Cottage, Auchleven, Insch, Aberdeenshire AB52 6QD (GB).

(74) Common Representative: **MERK PATENT GMBH**; Frankfurter Strasse 250, 64293 Darmstadt (DE).

(81) Designated States *(national)*: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW.

(84) Designated States *(regional)*: ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:
— *with international search report*

(88) Date of publication of the international search report:
11 December 2003

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

WO 02/077034 A3

(54) Title: MODIFIED GRANULOCYTE COLONY STIMULATING FACTOR (G-CSF) WITH REDUCED IMMUNOGENICITY

(57) Abstract: The present invention relates to polypeptides to be administered especially to humans and in particular for therapeutic use. The polypeptides are modified polypeptides whereby the modification results in a reduced propensity for the polypeptide to elicit an immune response upon administration to the human subject. The invention in particular relates to the modification of granulocyte colony stimulating factor (G-CSF) to result in granulocyte colony stimulating factor (G-CSF) proteins that are substantially non-immunogenic or less immunogenic than any non-modified counterpart when used *in vivo*.

INTERNATIONAL SEARCH REPORT

Inte al Application No
PCT/EP 02/01171

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 C07K14/535 A61K38/18 C12N15/27 C07K7/06 C07K7/08

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 7 C07K C12N A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

EPO-Internal, WPI Data, PAJ, BIOSIS

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	US 5 824 784 A (DEPRINCE RANDOLPH B ET AL) 20 October 1998 (1998-10-20) the whole document	1
X	EP 0 272 703 A (KYOWA HAKKO KOGYO KK) 29 June 1988 (1988-06-29) example 4	1-25
X	WO 00 34317 A (ADAIR FIONA SUZANNE ;CARR FRANCIS JOSEPH (GB); HAMILTON ANITA ANNE) 15 June 2000 (2000-06-15) the whole document	1-25
X	WO 98 52976 A (BIOVATION LTD ;CARR FRANCIS J (GB)) 26 November 1998 (1998-11-26) the whole document	1-25

-/--

[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

° 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 when the document is taken alone

"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

Date of the actual completion of the international search	Date of mailing of the international search report
26 July 2002	22.05.03

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	Friedrich, C

INTERNATIONAL SEARCH REPORT

Inte ıal Application No
PCT/EP 02/01171

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 98 59244 A (BROOKS TONY ;ECLAGEN LTD (GB); KEMP GRAHAM (GB); CARR FRANK (GB);) 30 December 1998 (1998-12-30) the whole document	1-25
A	LARICCHIA-ROBBIO LEOPOLDO ET AL: "Naturally occurring and therapy-induced antibodies to human granulocyte colony-stimulating factor (G-CSF) in human serum." JOURNAL OF CELLULAR PHYSIOLOGY, vol. 173, no. 2, November 1997 (1997-11), pages 219-226, XP002206160 ISSN: 0021-9541 the whole document	1-25
A	SALI A ET AL: "COMPARATIVE PROTEIN MODELLING BY SATISFACTION OF SPATIAL RESTRAINTS" JOURNAL OF MOLECULAR BIOLOGY, LONDON, GB, vol. 234, 1993, pages 779-815, XP002948615 ISSN: 0022-2836 the whole document	1-25
A	ALTUVIA Y ET AL: "RANKING POTENTIAL BINDING PEPTIDES TO MHC MOLECULES BY A COMPUTATIONAL THREADING APPROACH" JOURNAL OF MOLECULAR BIOLOGY, LONDON, GB, vol. 249, no. 1, 1995, pages 244-250, XP000925520 ISSN: 0022-2836 the whole document	1-25
A	BRUSIC VLADIMIR ET AL: "MHCPEP, a database of MHC-binding peptides: Update 1997." NUCLEIC ACIDS RESEARCH, vol. 26, no. 1, 1 January 1998 (1998-01-01), pages 368-371, XP002204283 ISSN: 0305-1048 the whole document	1-25
A	BOEHM H-J: "PREDICTION OF BINDING CONSTANTS OF PROTEIN LIGANDS: A FAST METHOD FOR THE PRIORITIZATION OF HITS OBTAINED FROM DE NOVO DESIGN OR 3D DATABASE SEARCH PROGRAMS" JOURNAL OF COMPUTER-AIDED MOLECULAR DESIGN, ESCOM SCIENCE PUBLISHERS BV, XX, vol. 4, no. 12, July 1998 (1998-07), pages 309-323, XP008004781 ISSN: 0920-654X the whole document	1-25

INTERNATIONAL SEARCH REPORT

tional application No.
PCT/EP 02/01171

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

2. ☐ Claims Nos.:
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. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

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

see additional sheet

1. ☐ As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.

2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. ☐ As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:

4. ☒ 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 claims Nos.:

1-25 in part

Remark on Protest

☐ The additional search fees were accompanied by the applicant's protest.

☐ No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

Invention 1: claims 1-25

G-CSF with leucine at position 3 substituted by alanine, a pharmaceutical composition comprising said modified G-CSF, and the method for its manufacturing

Inventions 2-13 claims 1-25 in part

G-CSF with leucine at position 3 substituted by cysteine (invention 2), aspartic acid (invention 3), glutamic acid (invention 4), glycine (invention 5), histidine (invention 6), lysine (invention 7), asparagine (invention 8), proline (invention 9), glutamine (invention 10), arginine (invention 11), serine (invention 12), or threonine (invention 13), a pharmaceutical composition comprising said modified G-CSF, and the method for its manufacturing

Inventions 14-63: claims 1-25 in part

G-CSF with a modification at position 9 (invention 14), 14 (invention 15), 15 (invention 15), 18 (invention 16), 21 (invention 17), etc. according to table 2 in the description, a pharmaceutical composition comprising said modified G-CSF, and the method for its manufacturing

Inventions (further): claims 1-25 in part

All combinations of modifications at more than one position as referred to in claims 7, 10-12, and 15.

Inventions (further): claims 26-29

Each 13mer as disclosed in table 1 of the description (51 inventions).

INTERNATIONAL SEARCH REPORT

ormation on patent family members

Int nal Application No
PCT/EP 02/01171

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
US 5824784 A	20-10-1998	AT 179991 T	15-05-1999
		AU 706700 B2	24-06-1999
		AU 1841995 A	06-05-1996
		AU 4887099 A	11-11-1999
		CA 2178752 A1	25-04-1996
		CA 2307142 A1	25-04-1996
		CN 1313343 A	19-09-2001
		CN 1139932 A ,B	08-01-1997
		DE 69509628 D1	17-06-1999
		DE 69509628 T2	16-09-1999
		DK 733067 T3	01-11-1999
		EP 0733067 A1	25-09-1996
		EP 0822199 A2	04-02-1998
		ES 2131811 T3	01-08-1999
		GR 3030526 T3	29-10-1999
		HK 1008826 A1	31-03-2000
		IL 112585 A	31-08-2000
		JP 11310600 A	09-11-1999
		JP 3177449 B2	18-06-2001
		JP 9025298 A	28-01-1997
		JP 9506116 T	17-06-1997
		JP 3177251 B2	18-06-2001
		KR 248111 B1	15-03-2000
		KR 261030 B1	15-06-2000
		NZ 281469 A	24-04-1997
		WO 9611953 A1	25-04-1996
		US 5985265 A	16-11-1999
		ZA 9501008 A	18-10-1996
EP 0272703 A	29-06-1988	DE 3786767 D1	02-09-1993
		DE 3786767 T2	13-01-1994
		DK 680987 A	24-06-1988
		EP 0272703 A1	29-06-1988
		ES 2059354 T3	16-11-1994
		HK 164195 A	27-10-1995
		JP 2766798 B2	18-06-1998
		JP 10052281 A	24-02-1998
		JP 2673099 B2	05-11-1997
		JP 7149798 A	13-06-1995
		JP 2628961 B2	09-07-1997
		JP 6092994 A	05-04-1994
		JP 6008317 B	02-02-1994
		JP 63267292 A	04-11-1988
		KR 9616560 B1	14-12-1996
		NO 875378 A ,B,	24-06-1988
		NO 940495 A	24-06-1988
		PT 86465 A ,B	01-01-1988
		US 6027720 A	22-02-2000
		US 5194592 A	16-03-1993
		US 5214132 A	25-05-1993
		US 5714581 A	03-02-1998
		US 5681720 A	28-10-1997
		US 5795968 A	18-08-1998
		US 5994518 A	30-11-1999
		US 5362853 A	08-11-1994
WO 0034317 A	15-06-2000	AU 1667600 A	26-06-2000
		CA 2342967 A1	15-06-2000

INTERNATIONAL SEARCH REPORT

ırmation on patent family members

Int onal Application No
PCT/EP 02/01171

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 0034317 A		CN 1294596 T	09-05-2001
		EP 1051432 A2	15-11-2000
		WO 0034317 A2	15-06-2000
		JP 2002534959 T	22-10-2002
		AU 4635699 A	24-01-2000
WO 9852976 A	26-11-1998	AU 736549 B2	02-08-2001
		AU 7539398 A	11-12-1998
		EP 0983303 A1	08-03-2000
		WO 9852976 A1	26-11-1998
		GB 2339430 A	26-01-2000
		JP 2002512624 T	23-04-2002
WO 9859244 A	30-12-1998	AU 8118398 A	04-01-1999
		WO 9859244 A1	30-12-1998