



(11) **EP 2 287 174 B8**

(12) **CORRECTED EUROPEAN PATENT SPECIFICATION**

(15) Correction information:
Corrected version no 1 (W1 B1)
Corrections, see
Bibliography INID code(s) 54

(51) Int Cl.:
C07K 1/16 (2006.01) **C12N 15/10** (2006.01)

(48) Corrigendum issued on:
07.12.2016 Bulletin 2016/49

(45) Date of publication and mention
of the grant of the patent:
27.07.2016 Bulletin 2016/30

(21) Application number: **10178856.0**

(22) Date of filing: **16.01.2009**

(54) **Enhanced purification of antibody fragments by apatite chromatography**

Verbesserte reinigung von antikörperfragmenten durch apatit-chromatographie

Purification améliorée de fragments d'anticorps par chromatographie sur hydroxyapatite

(84) Designated Contracting States:
AT BE BG CH CY CZ DE DK EE ES FI FR GB GR
HR HU IE IS IT LI LT LU LV MC MK MT NL NO PL
PT RO SE SI SK TR

(30) Priority: **24.07.2008 US 135787 P**
28.01.2008 US 62663 P
18.01.2008 US 11513 P
19.03.2008 US 69859 P
27.03.2008 US 70841 P
20.08.2008 US 189467 P

(43) Date of publication of application:
23.02.2011 Bulletin 2011/08

(62) Document number(s) of the earlier application(s) in
accordance with Art. 76 EPC:
09701572.1 / 2 242 762

(73) Proprietor: **Bio-Rad Laboratories, Inc.**
Hercules, CA 94547 (US)

(72) Inventor: **Gagnon, Peter S.**
Singapore 189647 (SG)

(74) Representative: **dompatent von Kreisler Selting**
Werner -
Partnerschaft von Patent- und Rechtsanwälten
mbB
Deichmannhaus am Dom
Bahnhofsvorplatz 1
50667 Köln (DE)

(56) References cited:
EP-A1- 0 256 836 EP-A1- 1 081 221
WO-A1-89/12678 WO-A2-03/059935

- **GORBUNOFF ET AL: "The interaction of proteins with hydroxyapatite - I. Role of protein charge and structure", ANALYTICAL BIOCHEMISTRY, ACADEMIC PRESS INC. NEW YORK, vol. 136, no. 2, 1 February 1984 (1984-02-01), pages 425-432, XP024825292, ISSN: 0003-2697 [retrieved on 1984-02-01]**
- **GORBUNOFF M J: 'The interaction of proteins with hydroxyapatite. II. Role of acidic and basic groups.' ANALYTICAL BIOCHEMISTRY FEB 1984 LNKD-PUBMED:6721143 vol. 136, no. 2, February 1984, pages 433 - 439, XP024825293 ISSN: 0003-2697**

Note: Within nine months of the publication of the mention of the grant of the European patent in the European Patent Bulletin, any person may give notice to the European Patent Office of opposition to that patent, in accordance with the Implementing Regulations. Notice of opposition shall not be deemed to have been filed until the opposition fee has been paid. (Art. 99(1) European Patent Convention).

EP 2 287 174 B8

- GORBUNOFF M J ET AL: 'The interaction of proteins with hydroxyapatite. III. Mechanism.' ANALYTICAL BIOCHEMISTRY FEB 1984 LNKD-PUBMED:6721144 vol. 136, no. 2, February 1984, pages 440 - 445, XP024825294 ISSN: 0003-2697
- GORBUNOFF M J: 'Protein chromatography on hydroxyapatite columns.' METHODS IN ENZYMOLOGY 1990 LNKD- PUBMED:2156129 vol. 182, 1990, pages 329 - 339, XP009057845 ISSN: 0076-6879
- HAKAMATSUKA TAKASHI ET AL: 'Purification of 2-hydroxyisoflavanone dehydratase from the cell cultures of Pueraria lobata' PHYTOCHEMISTRY (OXFORD) vol. 49, no. 2, September 1998, pages 497 - 505, XP004290126 ISSN: 0031-9422
- Gagnon Pete: "Phosphate-free buffer system a new frontier for apatite chromatography", , 4 May 2008 (2008-05-04), XP002610605, Retrieved from the Internet:
URL:<http://www.validated.com/revalbio/pdf/files/CHT.Frontier.pdf> [retrieved on 2010-11-22]
- GAGNON PETE ET AL: "Reverse calcium affinity purification of Fab with calcium derivatized hydroxyapatite.", JOURNAL OF IMMUNOLOGICAL METHODS 15 MAR 2009 LNKD- PUBMED:19150352, vol. 342, no. 1-2, 14 January 2009 (2009-01-14), pages 115-118, XP002610666, ISSN: 1872-7905
- GAGNON PETE: "Monoclonal antibody purification with hydroxyapatite.", NEW BIOTECHNOLOGY JUN 2009 LNKD-PUBMED:19491046, vol. 25, no. 5, June 2009 (2009-06), pages 287-293, XP002610667, ISSN: 1871-6784