



(51) International Patent Classification:

G01N33/58 (2006.01) G01N33/66 (2006.01)  
G01N21/64 (2006.01)

(21) International Application Number:

PCT/US20 12/0465 13

(22) International Filing Date:

12 July 2012 (12.07.2012)

(25) Filing Language:

English

(26) Publication Language:

English

(30) Priority Data:

61/508,509 15 July 2011 (15.07.2011) US

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(81) Designated States (unless otherwise indicated, for every kind of national protection available): AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN,

HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

(84) Designated States (unless otherwise indicated, for every kind of regional protection available): ARIPO (BW, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, RU, TJ, TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Declarations under Rule 4.17:

- as to applicant's entitlement to apply for and be granted a patent (Rule 4.17(H))
- as to the applicant's entitlement to claim the priority of the earlier application (Rule 4.17(in))
- of inventorship (Rule 4.17(iv))

Published:

- with international search report (Art. 21(3))
- before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments (Rule 48.2(h))

(88) Date of publication of the international search report:

20 June 2013

(54) Title: FLUOROPHORES AND THEIR COMBINATION WITH PYRIDINIUM BORONIC ACID QUENCHERS USE IN ANALYTE SENSORS

(57) Abstract: Aspects of the invention relate generally to the detection of polyhydroxyl- substituted organic molecules, and in particular to the use of fluorescent dyes combined with pyridinium salts functionalized with boronic acids as quenchers for use in glucose sensors. The fluorescent dye is an anionic, non-pyrene sulfonate-based fluorescent dye that is excited by visible light and emits at a wavelength greater than 500 nm. The dye is functionalized with at least one reactive group and is, for example, selected from sulforhodamine B, sulforhodamine 101, carboxytetramethylrhodamine, tetrakis(4-sulfophenyl)porphine, tetrakis(4-carboxyphenyl)porphine, potassium perylene-tetracarboxylate, Alexa Fluor 532, Alexa Fluor 546, Alexa Fluor 568, Alexa Fluor 594, BODIPY 576/589, BODIPY 581/591, BODIPY TR-X, SNARF-1, and Texas Red.



# INTERNATIONAL SEARCH REPORT

International application No PCT/US2012/Q465 13
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**A. CLASSIFICATION OF SUBJECT MATTER**  
 I NV . G91N33/58 G0 1N2 1/64 GQ1N33/66  
 ADD .

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)  
 G01N

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 practicable, search terms used)  
 EPO-Internal , CHEM ABS Data , WPI Data

**C. DOCUMENTS CONSIDERED TO BE RELEVANT**

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	GUNTER ZENKL ET AL: "Sugar- Respon sive Fl uorescent Nanospheres " , MACROMOLECU LAR BIOSC I ENC E, vol . 8, no. 2, 11 February 2008 (2008-02 - 11) , pages 146- 152 , XP055046244 , I SSN : 1616-5 187 , DOI : 10.1002/mabi .200700174	1- 5 , 7 , 8
A	abstract ; f i g u r e s 2 - 4 ----- -/--	11 - 17

Further documents are listed in the continuation of Box C.       See patent family annex.

\* Special categories of cited documents :

<p>"A" document defining the general state of the art which is not considered to be of particular relevance</p> <p>"E" earlier application or patent but published on or after the international filing date</p> <p>"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)</p> <p>"O" document referring to an oral disclosure, use, exhibition or other means</p> <p>"P" document published prior to the international filing date but later than the priority date claimed</p>	<p>"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</p> <p>"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</p> <p>"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</p> <p>"&amp;" document member of the same patent family</p>
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Date of the actual completion of the international search  12 December 2012	Date of mailing of the international search report  08/05/2013
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Name and mailing address of the ISA/ European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016	Authorized officer  Fausti , Si mone
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## INTERNATIONAL SEARCH REPORT

International application No  
PCT/US2012/Q46513

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT		
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	HUA YANG ET AL: "Scalable Synthesis of Lissamine Rhodamine B Sulfonyl Chloride and Incorporation of Xanthene Derivatives onto Polymer Supports" , SYNTHESIS, vol . 2008, no. 6, 1 March 2008 [2008-03-01] , pages 957-961, XP055034973, ISSN: 0039-7881, DOI : 10. 1055/ s-2008-1032172 abstract; compounds 5,7,9 -----	1-5,7,8
Y	US 2009/061528 AI (SURI JEFF T [US] ) 5 March 2009 [2009-03-05] cited in the application paragraph [0078] ; claims 1-24,27-30; figures 1,4-7 -----	1-8, 11-17
X	CORDES DAVID B: "Optical glucose detection across the visible spectrum using anionic fluorescent dyes and a viologen quencher in a two-component saccharide sensing system" , ORGANIC & BIOMOLECULAR CHEMISTRY, ROYAL SOCIETY OF CHEMISTRY, GB, vol . 3, no. 9, 7 May 2005 (2005-05-07) , pages 1708-1713, XP008081119 , ISSN: 1477-0520, DOI : 10. 1039/B418953A abstract; figure 1 -----	1-3,8, 13-17
Y	-----	1-8, 11-17
A	US 2002/193672 AI (WALSH JOSEPH C [US] ET AL) 19 December 2002 (2002-12-19) cited in the application abstract; figures 3,6,7,9-12 , 15,16 -----	1-8, 11-17
A	DI WANG ET AL: "Stimuli-Responsive Fluorescent Poly( N -isopropyl acrylamide) Microgels Labeled with Phenylboronic Acid Moieties as Multifunctional Ratiometric Probes for Glucose and Temperatures" , MACROMOLECULES, vol . 44, no. 7, 12 April 2011 (2011-04-12) , pages 2282-2290, XP055046889, ISSN: 0024-9297, DOI : 10. 1021/ma200053a abstract; figures 3-4 -----	1-8, 11-17
Y	US 2009/081803 AI (GAMSEY SOYA [US] ET AL) 26 March 2009 (2009-03-26) cited in the application claims 3,7,22-24 -----	15,16
Y	US 2008/305009 AI (GAMSEY SOYA [US] ET AL) 11 December 2008 (2008-12-11) cited in the application claims 1,3,5 , 10,12, 15,20,23,29,30 -----	15,16

# INTERNATIONAL SEARCH REPORT

International application No.  
PCT/US2012/046513

## Box No. II Observations where certain claims were found unsearchable (Continuation of item 2 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 64(a).

## Box No. III Observations where unity of invention is lacking (Continuation of item 3 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 fees, this Authority did not invite payment of additional fees.
  
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.:  
11, 12(completely) ; 1-8, 13-17 (partially)

### Remark on Protest

- The additional search fees were accompanied by the applicant's protest and, where applicable, the payment of a protest fee.
- The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation.
- No protest accompanied the payment of additional search fees.

# INTERNATIONAL SEARCH REPORT

Information on patent Family members

International application No PCT/US2012/Q46513
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Patent document cited in search report	Publication date	Patent family member(s)	Publication date
US 2009061528	AI	05-03-2009	EP 2181160 AI 05-05-2010
			JP 2010535903 A 25-11-2010
			US 2009061528 AI 05-03-2009
			WO 2009021057 AI 12-02-2009
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US 2002193672	AI	19-12-2002	NONE
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US 2009081803	AI	26-03-2009	EP 2222686 A2 01-09-2010
			JP 2011511755 A 14-04-2011
			US 2009081803 AI 26-03-2009
			WO 2009009756 A2 15-01-2009
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US 2008305009	AI	11-12-2008	AT 506368 T 15-05-2011
			CA 2684511 AI 13-11-2008
			EP 2147003 AI 27-01-2010
			JP 2010526094 A 29-07-2010
			US 2008305009 AI 11-12-2008
			US 2011171742 AI 14-07-2011
			US 2012208286 AI 16-08-2012
			WO 2008137604 AI 13-11-2008
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This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

1. claims: 11, 12(completely) ; 1-8, 13-17(parti ally)

fluorescent dyes, which contain the sulforhodamine B fluorophore and are functional ized with reactive groups, e.g. sulforhodamine B CysMA and the syntheti c method for its preparati on, as well as analyte sensors and devi ces compri sing (any of) these dyes.

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2. claims: 1-10, 13-17(al l parti ally)

fluorescent dyes, which contain the sulforhodamine 101 (al ias Texas Red) fluorophore and are functional ized with reactive groups, syntheti c methods for thei r preparation , analyte sensors and devi ces compri sing (any of) these dyes

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3. claims: 1-10, 13-17(al l parti ally)

fluorescent dyes, which contain the carboxytetramethyl rhodamine fluorophore and are functional ized with reactive groups, syntheti c methods for thei r preparation, analyte sensors and devi ces compri sing (any of) these dyes

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4. claims: 1-10, 13-17(al l parti ally)

fluorescent dyes, which contain a porphine fluorophore and are functional ized with reactive groups, syntheti c methods for thei r preparation, analyte sensors and devi ces compri sing (any of) these dyes

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5. claims: 1-10, 13-17(al l parti ally)

fluorescent dyes, which contain a perylene fluorophore and are functional ized with reactive groups, syntheti c methods for thei r preparation, analyte sensors and devi ces compri sing (any of) these dyes

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6. claims: 1-10, 13-17(al l parti ally)

fluorescent dyes, which contain the Alexa Fluor 532 fluorophore and are functional ized with reactive groups, syntheti c methods for thei r preparati on, analyte sensors and devi ces compri sing (any of) these dyes

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7. claims: 1-10, 13-17(al l parti ally)

fluorescent dyes, which contain the Alexa Fluor 546 fluorophore and are functional ized with reactive groups, syntheti c methods for thei r preparati on, analyte sensors and devi ces compri sing (any of) these dyes  
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8. claims: 1-10, 13-17(al l parti al ly)

fluorescent dyes, which contain the Alexa Fluor 568 fluorophore and are functional ized with reactive groups, syntheti c methods for thei r preparati on, analyte sensors and devi ces compri sing (any of) these dyes  
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9. claims: 1-10, 13-17(al l parti al ly)

fluorescent dyes, which contain the Alexa Fluor 594 fluorophore and are functional ized with reactive groups, syntheti c methods for thei r preparati on, analyte sensors and devi ces compri sing (any of) these dyes  
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10. claims: 1-10, 13-17(al l partial ly)

fluorescent dyes, which contain a bodipy fluorophore and are functional ized with reactive groups, syntheti c methods for thei r preparation, analyte sensors and devi ces compri sing (any of) these dyes  
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11. claims: 1-10, 13-17(al l partial ly)

fluorescent dyes, which contain the Snarf-1 fluorophore and are functional ized with reactive groups, syntheti c methods for thei r preparation, analyte sensors and devi ces compri sing (any of) these dyes  
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