



INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

<p>(51) International Patent Classification ⁶ : C07D 471/04, 491/04, 501/18, C12Q 1/00</p>	<p>A3</p>	<p>(11) International Publication Number: WO 96/30540</p> <p>(43) International Publication Date: 3 October 1996 (03.10.96)</p>
<p>(21) International Application Number: PCT/US96/04059</p> <p>(22) International Filing Date: 20 March 1996 (20.03.96)</p> <p>(30) Priority Data: 08/407,544 20 March 1995 (20.03.95) US</p> <p>(60) Parent Application or Grant (63) Related by Continuation US 08/407,544 (CIP) Filed on 20 March 1995 (20.03.95)</p> <p>(71) Applicant (for all designated States except US): THE REGENTS OF THE UNIVERSITY OF CALIFORNIA [US/US]; 22nd floor, 300 Lakeside Drive, Oakland, CA 94612 (US).</p> <p>(72) Inventors; and (75) Inventors/Applicants (for US only): TSIEN, Roger, Y. [US/US]; 8535 Nottingham Place, La Jolla, CA 92037 (US). ZLOKARNIK, Gregor [DE/US]; 3329 Jemez Drive, San Diego, CA 92117 (US).</p>	<p>(74) Agent: STORELLA, John, R.; Townsend and Townsend and Crew, Steuart Street Tower, 20th floor, One Market Plaza, San Francisco, CA 94105 (US).</p> <p>(81) Designated States: AL, AM, AT, AU, AZ, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, EE, ES, FI, GB, GE, HU, IS, JP, KE, KG, KP, KR, KZ, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, TJ, TM, TR, TT, UA, UG, US, UZ, VN, ARIPO patent (KE, LS, MW, SD, SZ, UG), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG).</p> <p>Published <i>With international search report.</i> <i>Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.</i></p> <p>(88) Date of publication of the international search report: 9 January 1997 (09.01.97)</p>	
<p>(54) Title: SUBSTRATES FOR BETA-LACTAMASE AND USES THEREOF</p>		
<div style="text-align: center;"> <p style="text-align: right;">(I)</p> </div>		
<p>(57) Abstract</p> <p>Substrates for β-lactamase of general formula (I) in which one of X and Y is a fluorescent donor moiety and the other is a quencher (which may or may not re-emit); R' is selected from the group consisting of H, lower (i.e., alkyl of 1 to about 5 carbon atoms) and $(\text{CH}_2)_n\text{OH}$, in which n is 0 or an integer from 1 to 5; R'' is selected from the group consisting of H, physiologically acceptable metal and ammonium cations, $-\text{CHR}^2\text{OCO}(\text{CH}_2)_n\text{CH}_3$, $-\text{CHR}^2\text{OCOC}(\text{CH}_3)_3$, acylthiomethyl, acyloxy-alpha-benzyl, delta-butyrolactonyl, methoxycarbonyloxymethyl, phenyl, methylsulphinylmethyl, beta-morpholinoethyl, dialkylaminoethyl, acyloxyalkyl, dialkylaminocarbonyloxymethyl and aliphatic, in which R² is selected from the group consisting of H and lower alkyl; A is selected from the group consisting of S, O, SO, SO₂ and CH₂; and Z' and Z'' are linkers for the fluorescent donor and quencher moieties. Methods of assaying β-lactamase activity and monitoring expression in systems using β-lactamase as a reporter gene also are disclosed.</p>		

FOR THE PURPOSES OF INFORMATION ONLY

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AM	Armenia	GB	United Kingdom	MW	Malawi
AT	Austria	GE	Georgia	MX	Mexico
AU	Australia	GN	Guinea	NE	Niger
BB	Barbados	GR	Greece	NL	Netherlands
BE	Belgium	HU	Hungary	NO	Norway
BF	Burkina Faso	IE	Ireland	NZ	New Zealand
BG	Bulgaria	IT	Italy	PL	Poland
BJ	Benin	JP	Japan	PT	Portugal
BR	Brazil	KE	Kenya	RO	Romania
BY	Belarus	KG	Kyrgystan	RU	Russian Federation
CA	Canada	KP	Democratic People's Republic of Korea	SD	Sudan
CF	Central African Republic	KR	Republic of Korea	SE	Sweden
CG	Congo	KZ	Kazakhstan	SG	Singapore
CH	Switzerland	LI	Liechtenstein	SI	Slovenia
CI	Côte d'Ivoire	LT	Lithuania	SK	Slovakia
CM	Cameroon	LK	Sri Lanka	SN	Senegal
CN	China	LR	Liberia	SZ	Swaziland
CS	Czechoslovakia	LT	Lithuania	TD	Chad
CZ	Czech Republic	LU	Luxembourg	TG	Togo
DE	Germany	LV	Latvia	TJ	Tajikistan
DK	Denmark	MC	Monaco	TT	Trinidad and Tobago
EE	Estonia	MD	Republic of Moldova	UA	Ukraine
ES	Spain	MG	Madagascar	UG	Uganda
FI	Finland	ML	Mali	US	United States of America
FR	France	MN	Mongolia	UZ	Uzbekistan
GA	Gabon	MR	Mauritania	VN	Viet Nam

INTERNATIONAL SEARCH REPORT

International Application No

PCT/US 96/04059

A. CLASSIFICATION OF SUBJECT MATTER <p style="text-align: center;">C 07 D 471/04, C 07 D 491/04, C 07D 501/18, C 12 Q 1/00</p> <p>According to International Patent Classification (IPC) or to both national classification and IPC 6</p>				
B. FIELDS SEARCHED Minimum documentation searched (classification system followed by classification symbols) <p style="text-align: center;">C 07 D, C 12 Q</p>				
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) 				
C. DOCUMENTS CONSIDERED TO BE RELEVANT				
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.		
A	EP, A, 0 354 757 (BEECHAM GROUP PLC) 14 February 1990 (14.02.90), claims 1-10.	1-93		
A	US, A, 5 162 524 (FARINA et al.) 10 November 1992 (10.11.92), claims 1-12.	1-93		
A	EP, A, 0 053 541 (RHONE-POULENC IND.) 09 June 1982 (09.06.82), claims 1-4.	1-93		
<input type="checkbox"/> Further documents are listed in the continuation of box C.				
<input type="checkbox"/> Patent family members are listed in annex.				
* Special categories of cited documents :				
<table style="width: 100%; border: none;"> <tr> <td style="width: 50%; border: none; vertical-align: top;"> <p>"A" document defining the general state of the art which is not considered to be of particular relevance</p> <p>"E" earlier document 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> </td> <td style="width: 50%; border: none; vertical-align: top;"> <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>"&" document member of the same patent family</p> </td> </tr> </table>			<p>"A" document defining the general state of the art which is not considered to be of particular relevance</p> <p>"E" earlier document 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>"&" document member of the same patent family</p>
<p>"A" document defining the general state of the art which is not considered to be of particular relevance</p> <p>"E" earlier document 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>"&" document member of the same patent family</p>			
Date of the actual completion of the international search <p style="text-align: center;">17 October 1996</p>	Date of mailing of the international search report <p style="text-align: center;">05.11.96</p>			
Name and mailing address of the ISA 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	Authorized officer <p style="text-align: center;">BRUS e.h.</p>			

ANHANG

zum internationalen Recherchenbericht über die internationale Patentanmeldung Nr.

ANNEX

to the International Search Report to the International Patent Application No.

ANNEXE

au rapport de recherche international relatif à la demande de brevet international n°

PCT/US 96/04059 SAE 130932

In diesem Anhang sind die Mitglieder der Patentfamilien der im obengenannten internationalen Recherchenbericht angeführten Patentdokumente angegeben. Diese Angaben dienen nur zur Unterrichtung und erfolgen ohne Gewähr.

This Annex lists the patent family members relating to the patent documents cited in the above-mentioned international search report. The Office is in no way liable for these particulars which are given merely for the purpose of information.

La présente annexe indique les membres de la famille de brevets relatifs aux documents de brevets cités dans le rapport de recherche international visée ci-dessus. Les renseignements fournis sont donnés à titre indicatif et n'engagent pas la responsabilité de l'Office.

Table with 4 columns: In Recherchenbericht angeführtes Patentdokument, Datum der Veröffentlichung, Mitglied(er) der Patentfamilie, Datum der Veröffentlichung. It lists patent families for EP A2, US A, and EP A1 with various sub-offices and dates.