

# The United States of America



## The Director of the United States Patent and Trademark Office

*Has received an application for a patent for a new and useful invention. The title and description of the invention are enclosed. The requirements of law have been complied with, and it has been determined that a patent on the invention shall be granted under the law.*

*Therefore, this*

### United States Patent

*Grants to the person(s) having title to this patent the right to exclude others from making, using, offering for sale, or selling the invention throughout the United States of America or importing the invention into the United States of America, and if the invention is a process, of the right to exclude others from using, offering for sale or selling throughout the United States of America, or importing into the United States of America, products made by that process, for the term set forth in 35 U.S.C. 154(a)(2) or (c)(1), subject to the payment of maintenance fees as provided by 35 U.S.C. 41(b). See the Maintenance Fee Notice on the inside of the cover.*

*Michelle H. Lee*

*Director of the United States Patent and Trademark Office*



US009505976B2

(12) **United States Patent**  
**Lee et al.**

(10) **Patent No.:** **US 9,505,976 B2**

(45) **Date of Patent:** **Nov. 29, 2016**

(54) **EXCITATION-DEPENDENT FLUORESCENT ORGANIC NANOPARTICLES**

(71) Applicant: **National Kaohsiung Normal University**, Kaohsiung (TW)

(72) Inventors: **Kwang-Ming Lee**, Kaohsiung (TW);  
**Jing-Jong Shyue**, Kaohsiung (TW);  
**Sarah Y. Chang**, Kaohsiung (TW);  
**Thomas C. Yang**, Kaohsiung (TW)

(73) Assignee: **National Kaohsiung Normal University**, Kaohsiung (TW)

(\*) Notice: Subject to any disclaimer, the term of this patent is extended or adjusted under 35 U.S.C. 154(b) by 286 days.

(21) Appl. No.: **14/016,037**

(22) Filed: **Aug. 30, 2013**

(65) **Prior Publication Data**

US 2015/0064799 A1 Mar. 5, 2015

(51) **Int. Cl.**  
**G01N 21/64** (2006.01)  
**C09K 11/06** (2006.01)  
**B82Y 15/00** (2011.01)

(52) **U.S. Cl.**  
CPC ..... **C09K 11/06** (2013.01); **B82Y 15/00** (2013.01)

(58) **Field of Classification Search**  
CPC ..... C09K 11/06; B82Y 15/00  
See application file for complete search history.

(56) **References Cited**

FOREIGN PATENT DOCUMENTS

CN	102818790 A	12/2012
CN	103044581 A	4/2013
WO	WO 2012/109755 A1	8/2012

OTHER PUBLICATIONS

Lee et al., Excitation-dependent visible fluorescence in decameric nanoparticles with monoacylglycerol cluster chromophores, *Nature Communications* 4, 2013, Article No. 1544, pp. 1-8.\*  
Weng et al., Conformational dynamics and excitation wavelength dependent photoluminescence of decameric organic nanoparticles. *Phys. Chem. Chem. Phys.* 2013, vol. 15, pp. 16935-16940.\*  
H. Ohgi, et al., "Highly Isotactic Poly(Vinyl Alcohol) Derived From tert-Butyl Vinyl Ether," Part IV. Some Physical Properties, Structure and Hydrogen Bonding of Highly Isotactic Poly (Vinyl Alcohol) Films. *Polymer* 47, pp. 1324-1332 (2006).  
Marek Józefowicz et al, Excitation-wavelength Dependent Fluorescence of Ethyl 5-(4-aminophenyl)-3-amino-2,4-dicyanobenzoate, *J. Floresc.*, 2011, 21:239-245.

\* cited by examiner

*Primary Examiner* — Shafiqul Haq

(74) *Attorney, Agent, or Firm* — CKC & Partners Co., Ltd.

(57) **ABSTRACT**

A series of self-assembled monoacylglycerol clusters with excitation-dependent visible fluorescence have been developed.

**1 Claim, 29 Drawing Sheets**  
**(26 of 29 Drawing Sheet(s) Filed in Color)**

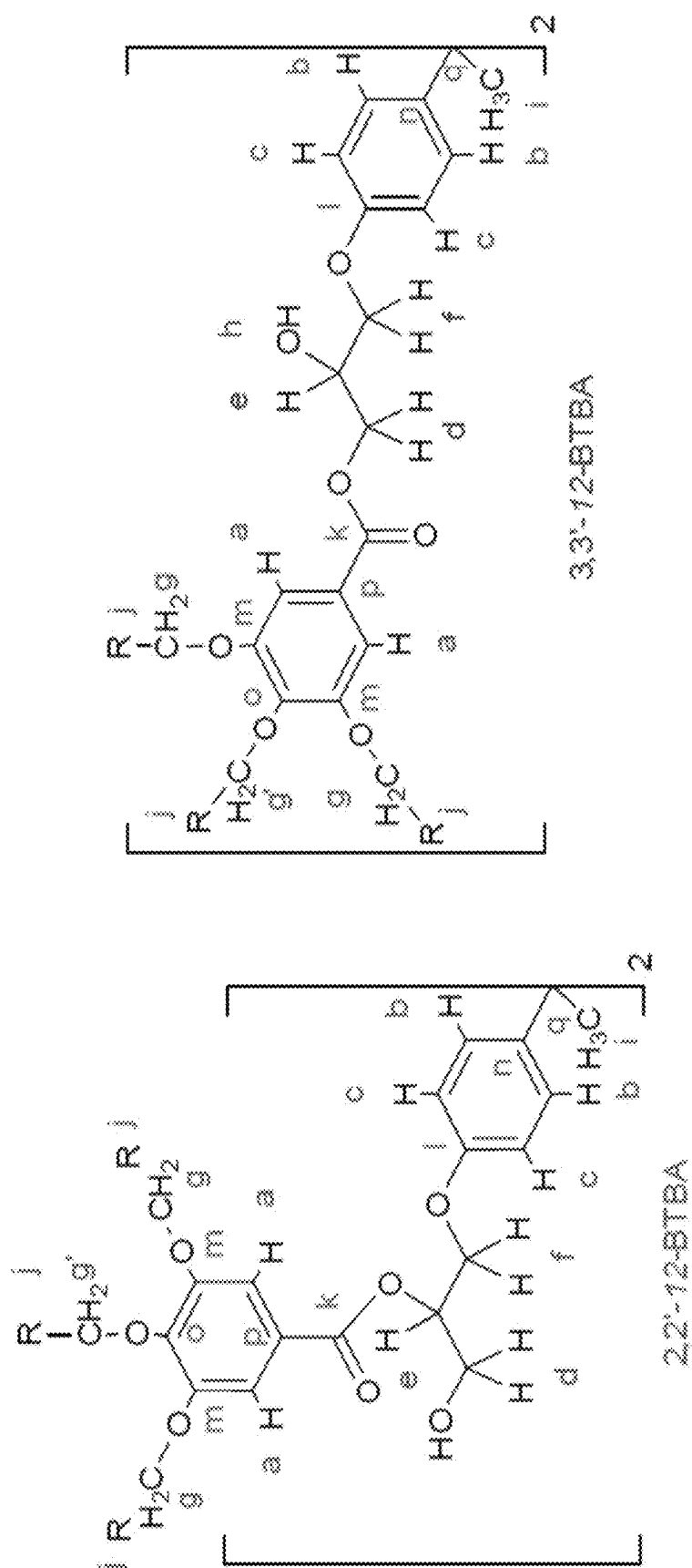


Fig. 1A

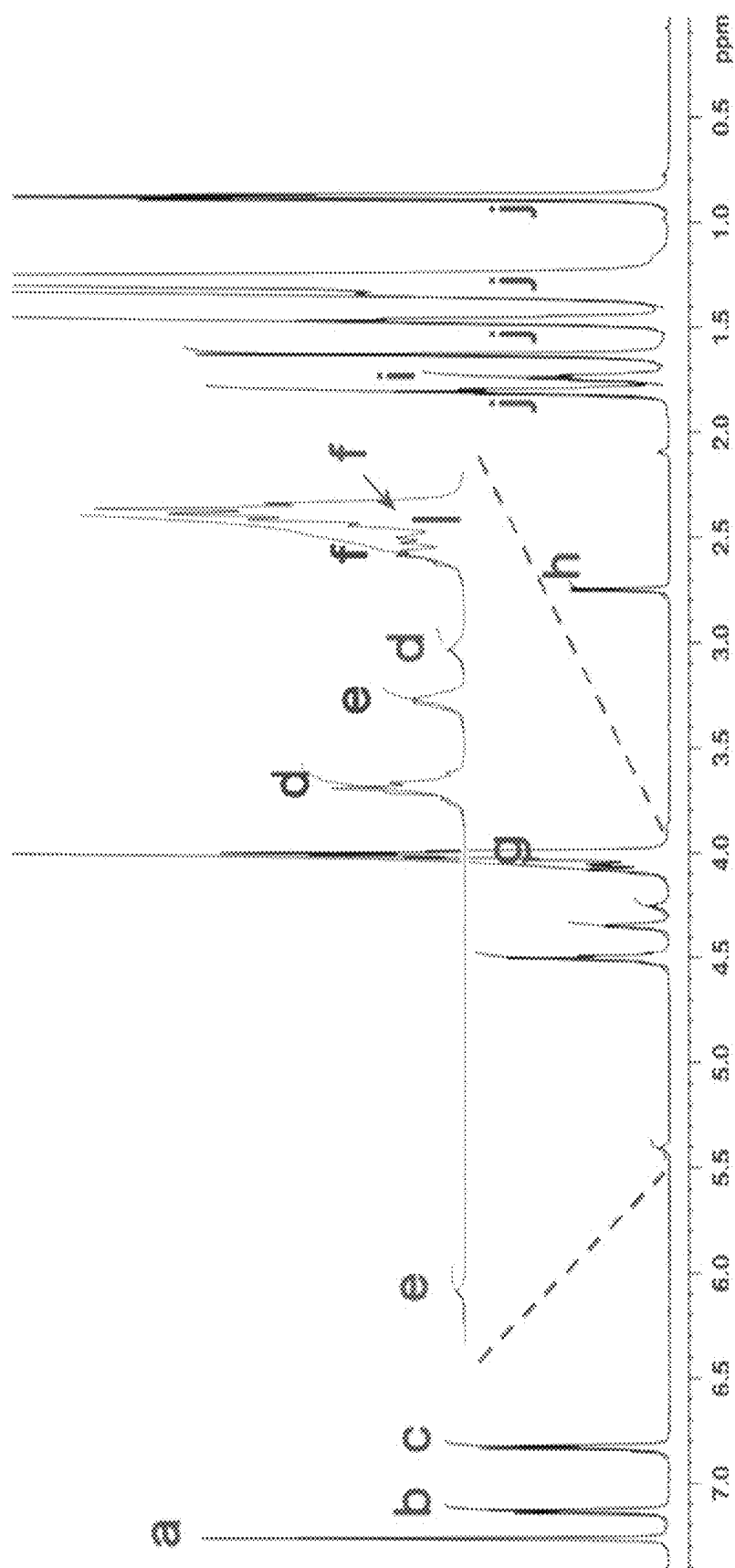


Fig. 1B

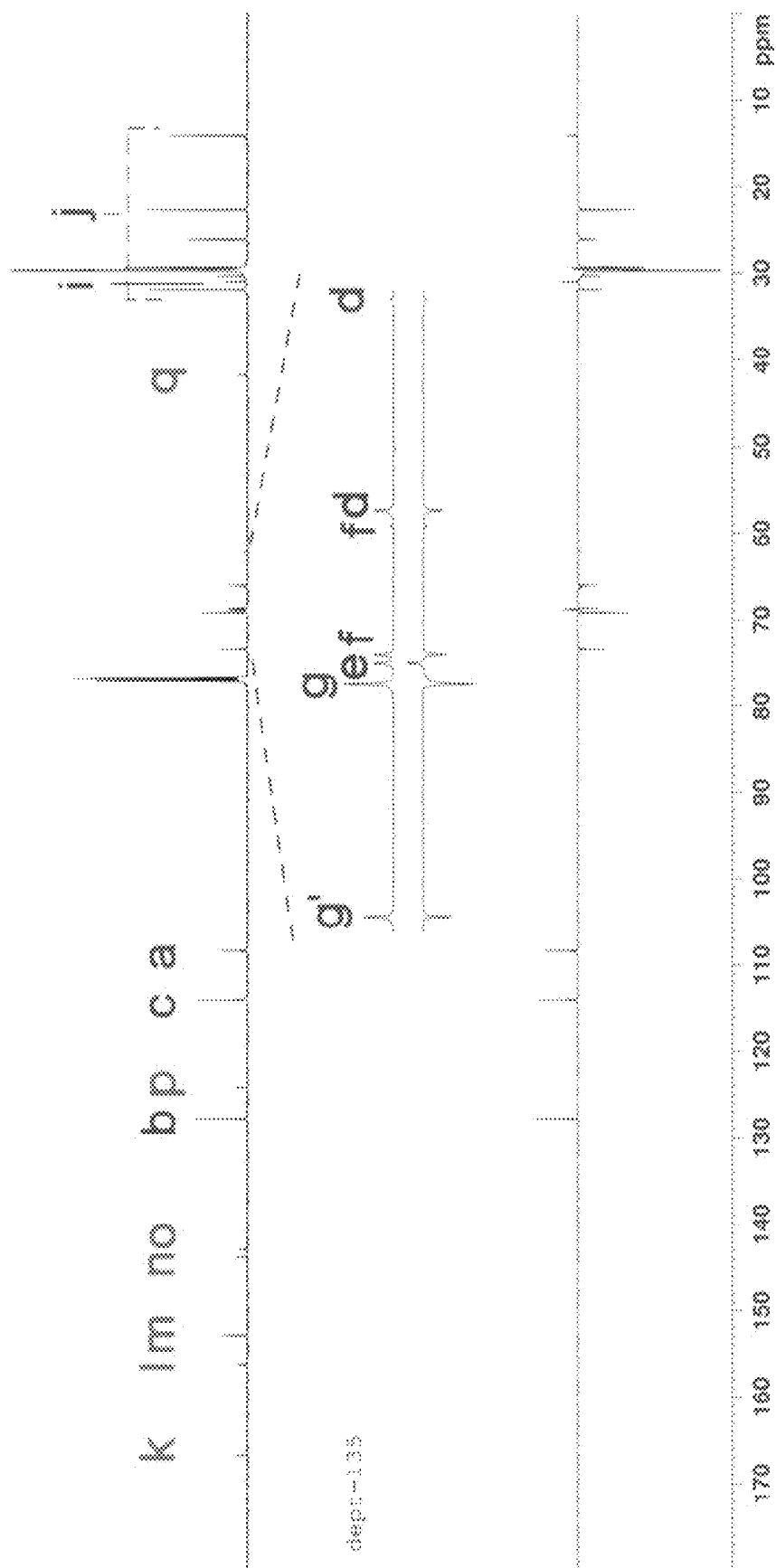


Fig. 1C

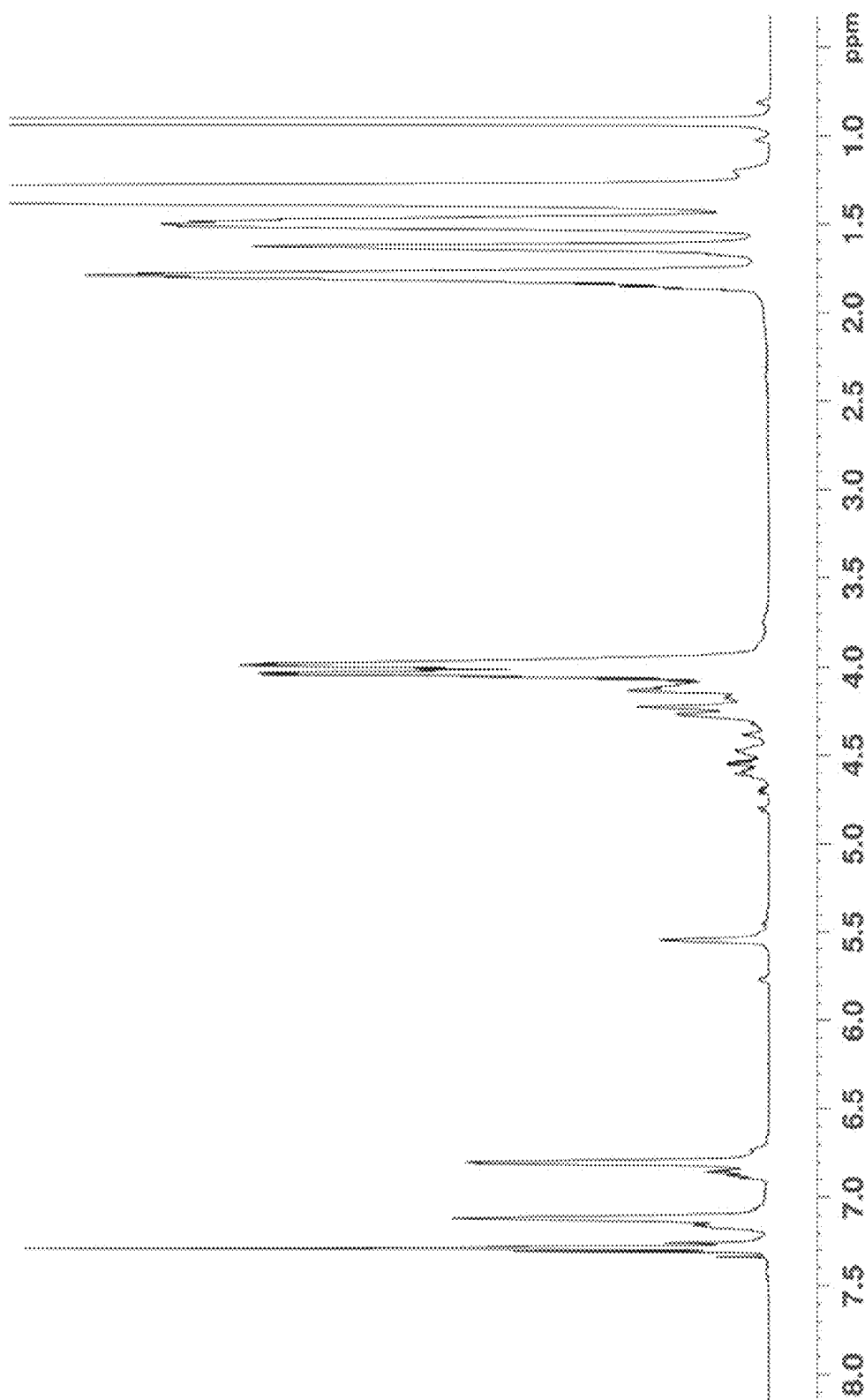


Fig. 2A

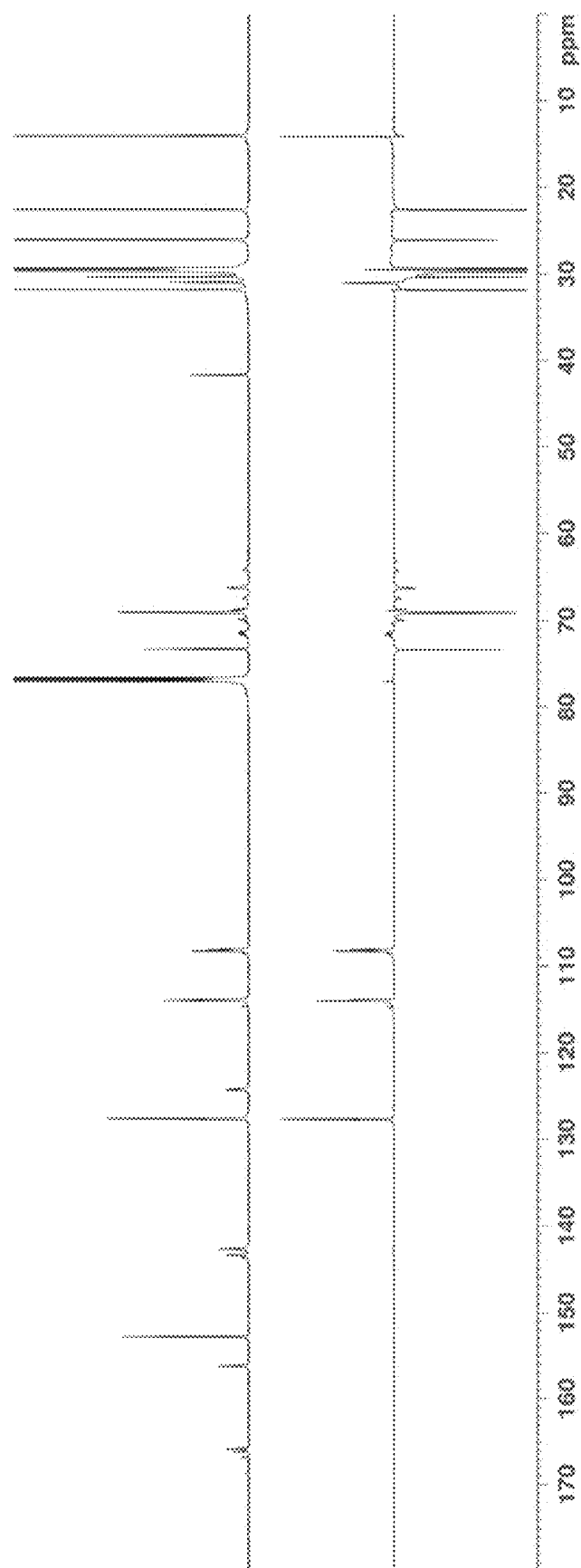


Fig. 2B

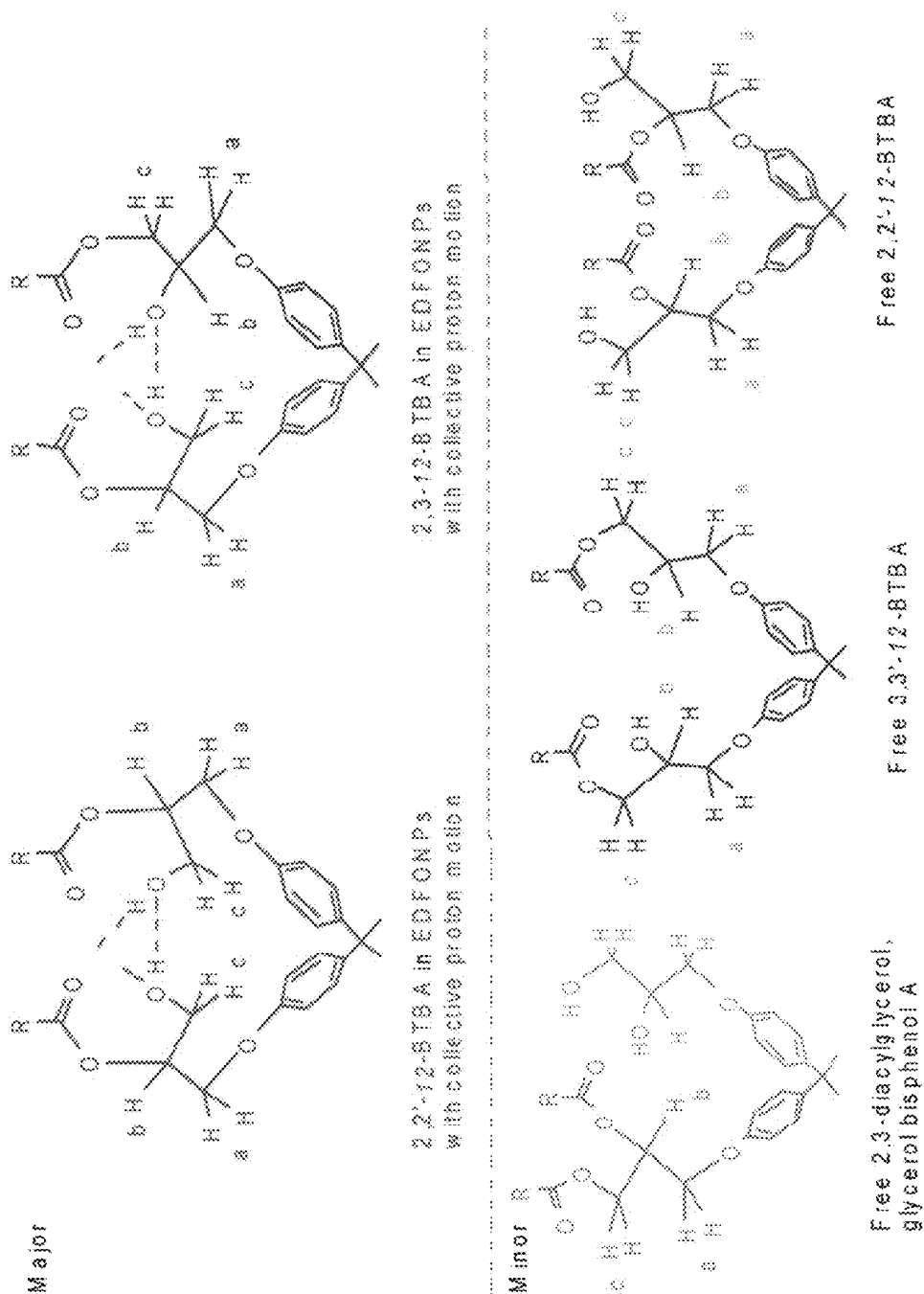


Fig. 2C

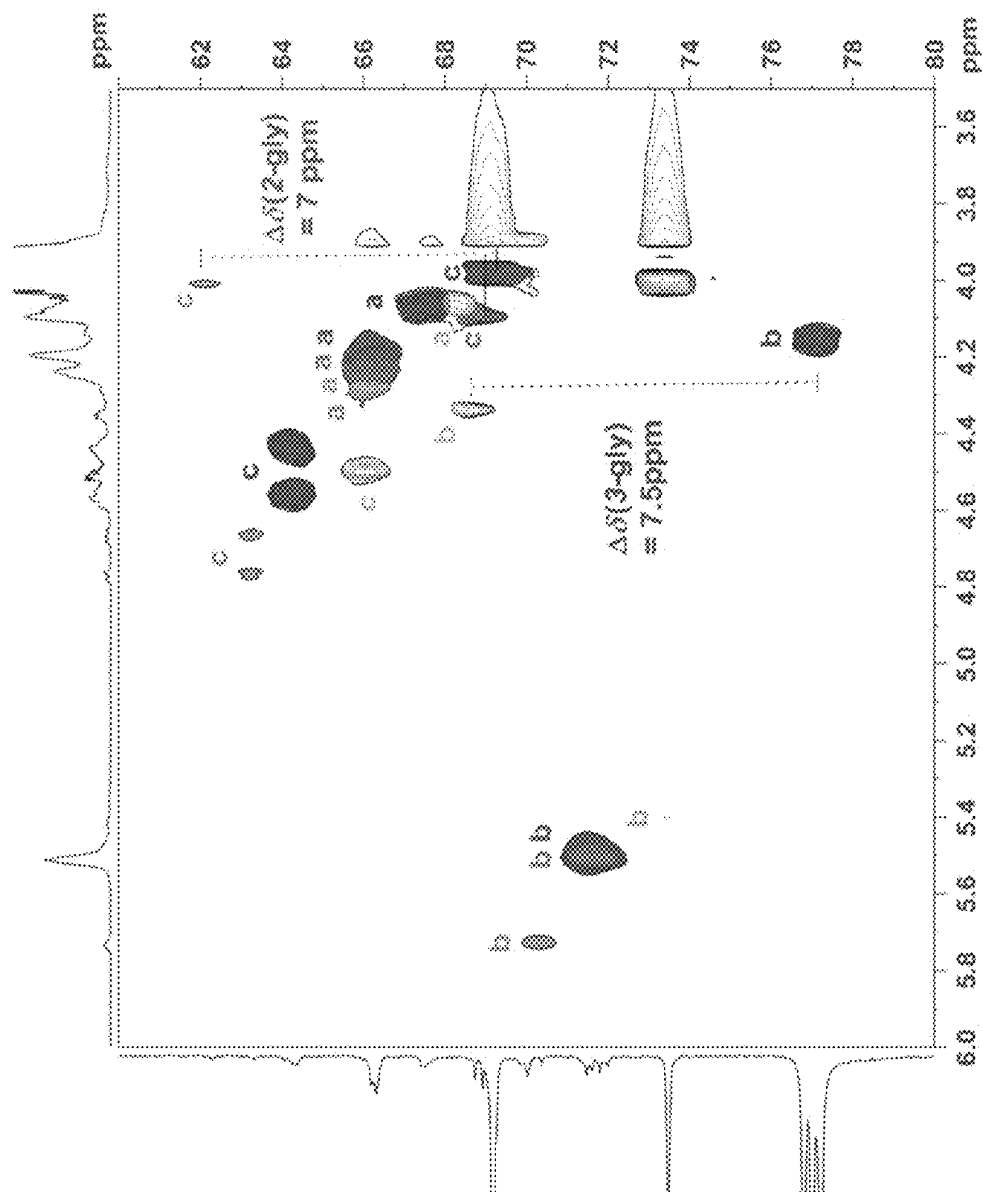
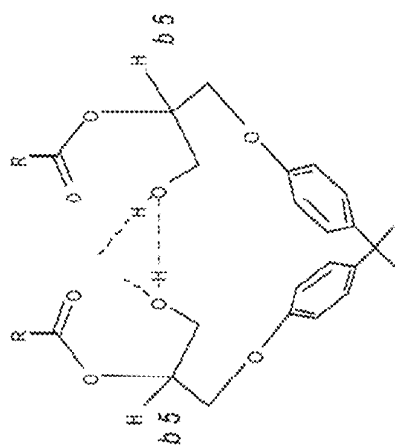
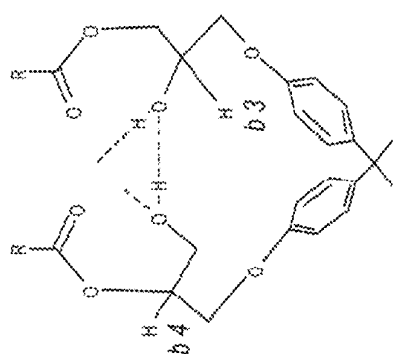


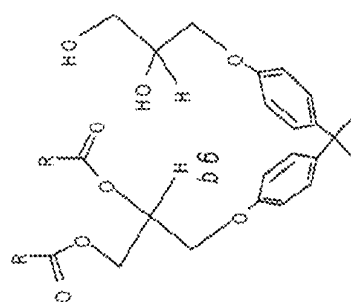
Fig. 2D



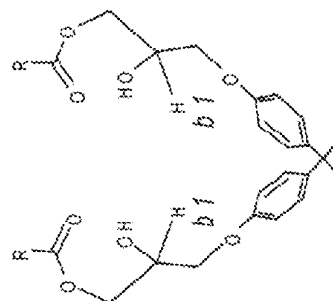
2,2'-12-BTBA in EDFONPs  
with collective proton motion



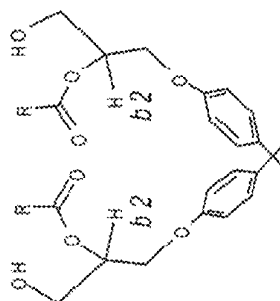
2,3-12-BTBA in EDFONPs  
with collective proton motion



Free 2,3-diacylglycerol,  
glycerol bisphenol A



Free 3,3'-12-BTBA



Free 2,2'-12-BTBA

Fig. 3A

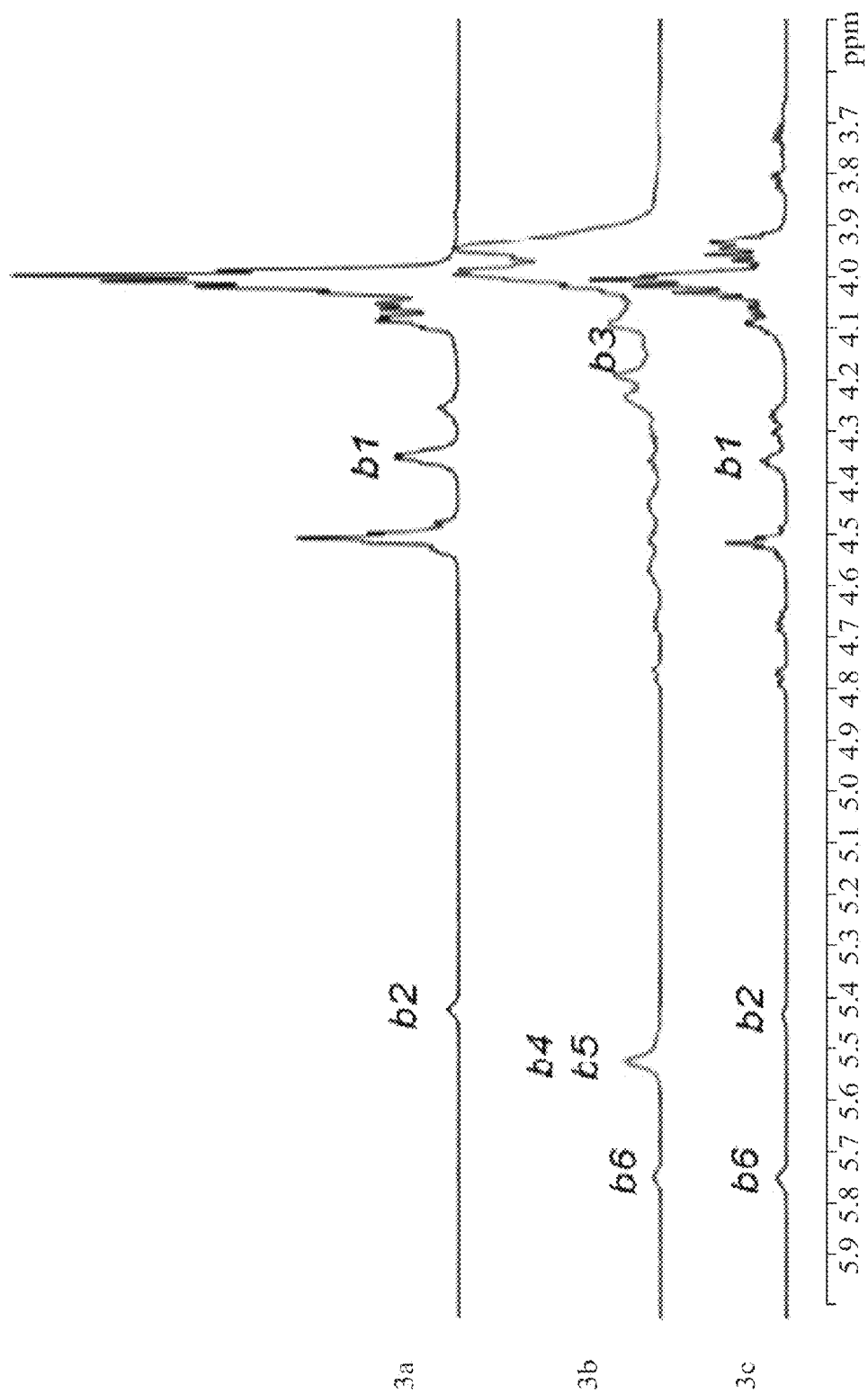


Fig. 3B

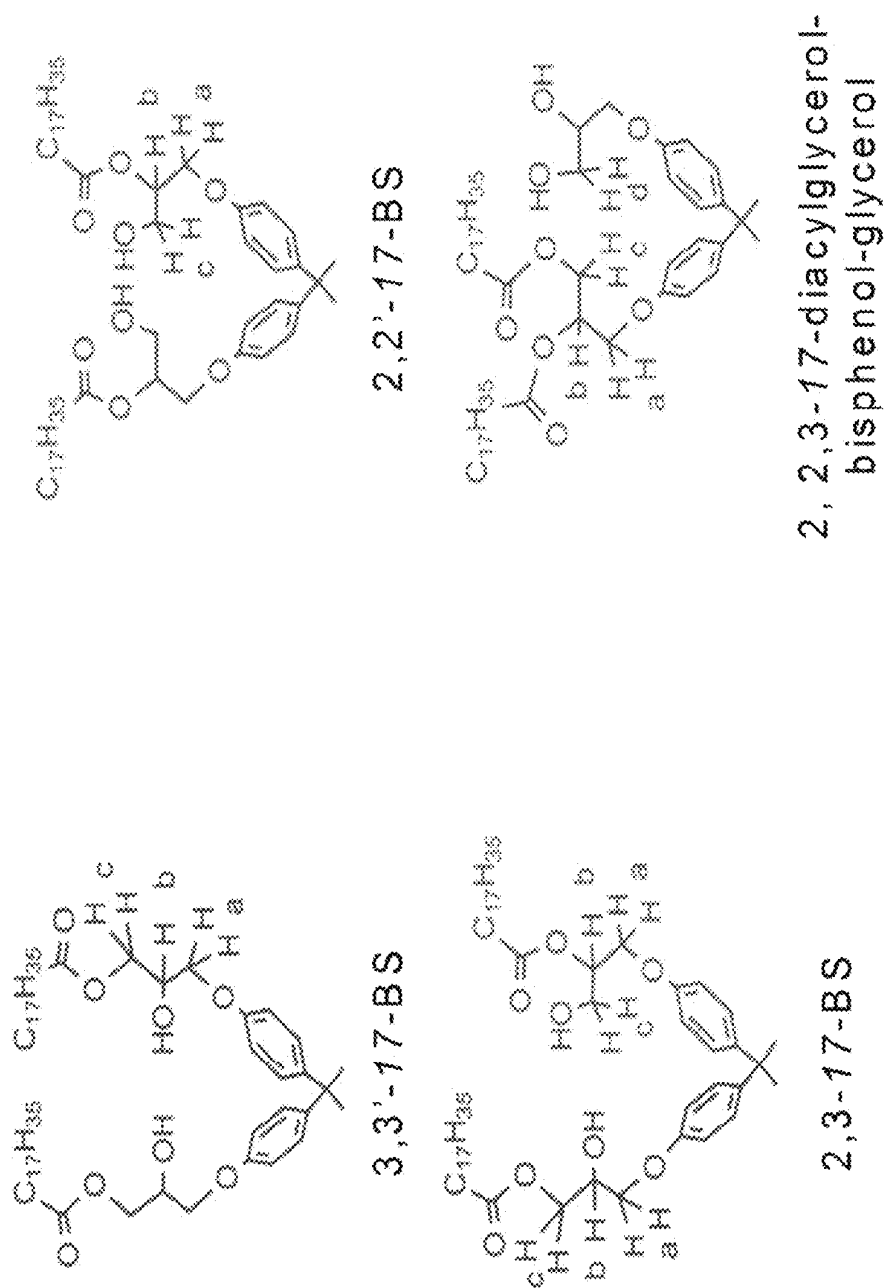


Fig. 4A

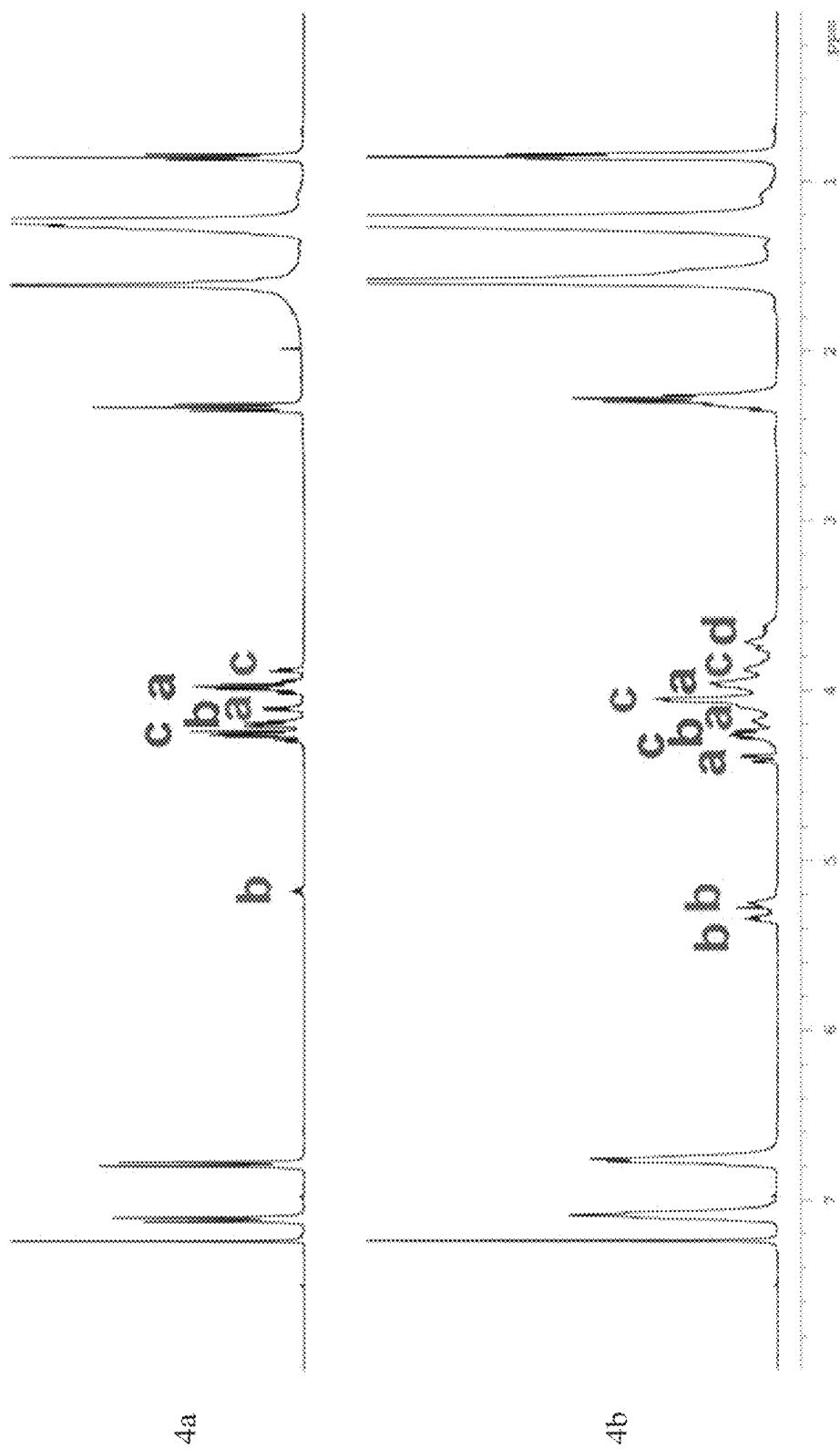


Fig. 4B

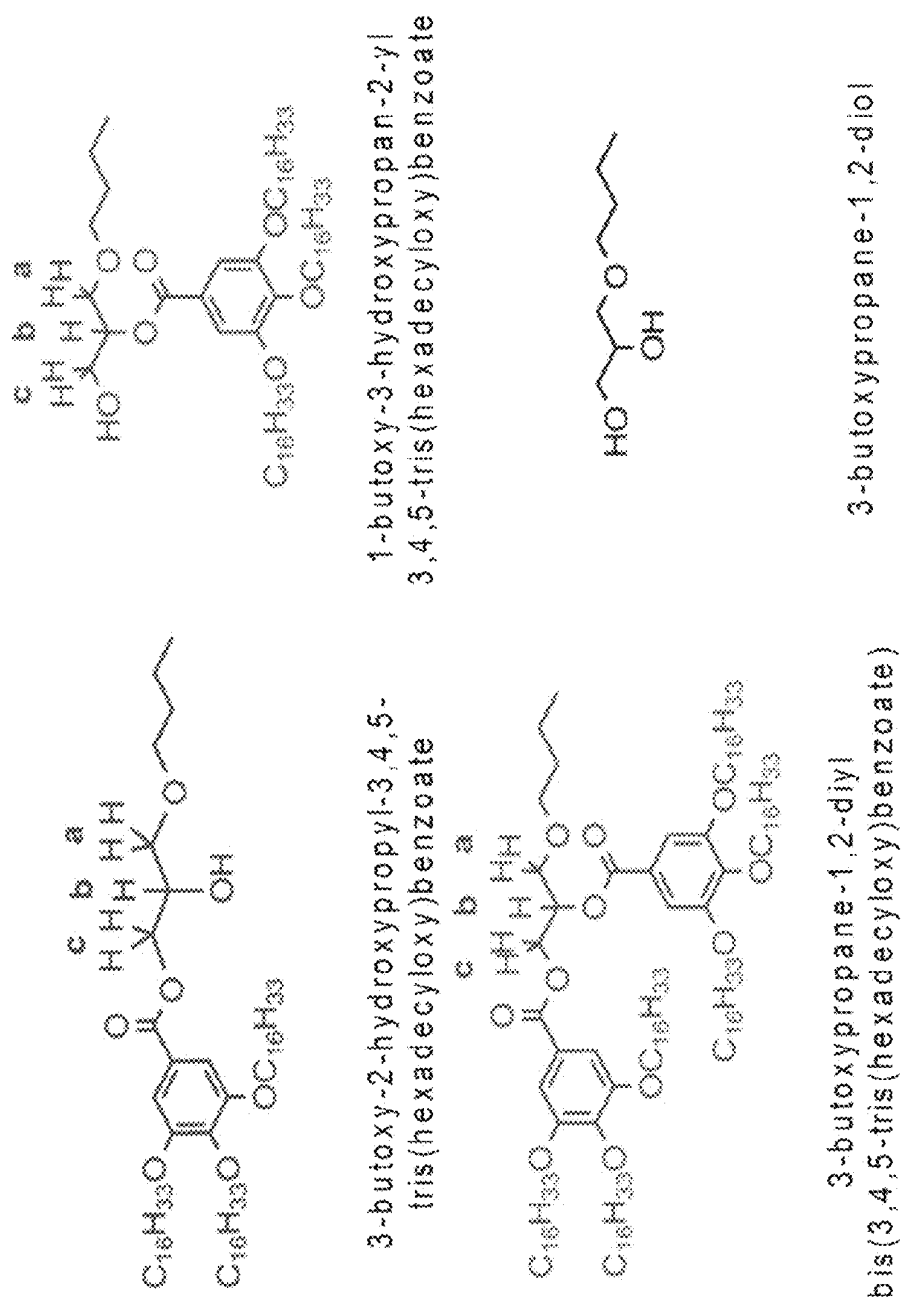


Fig. 5A

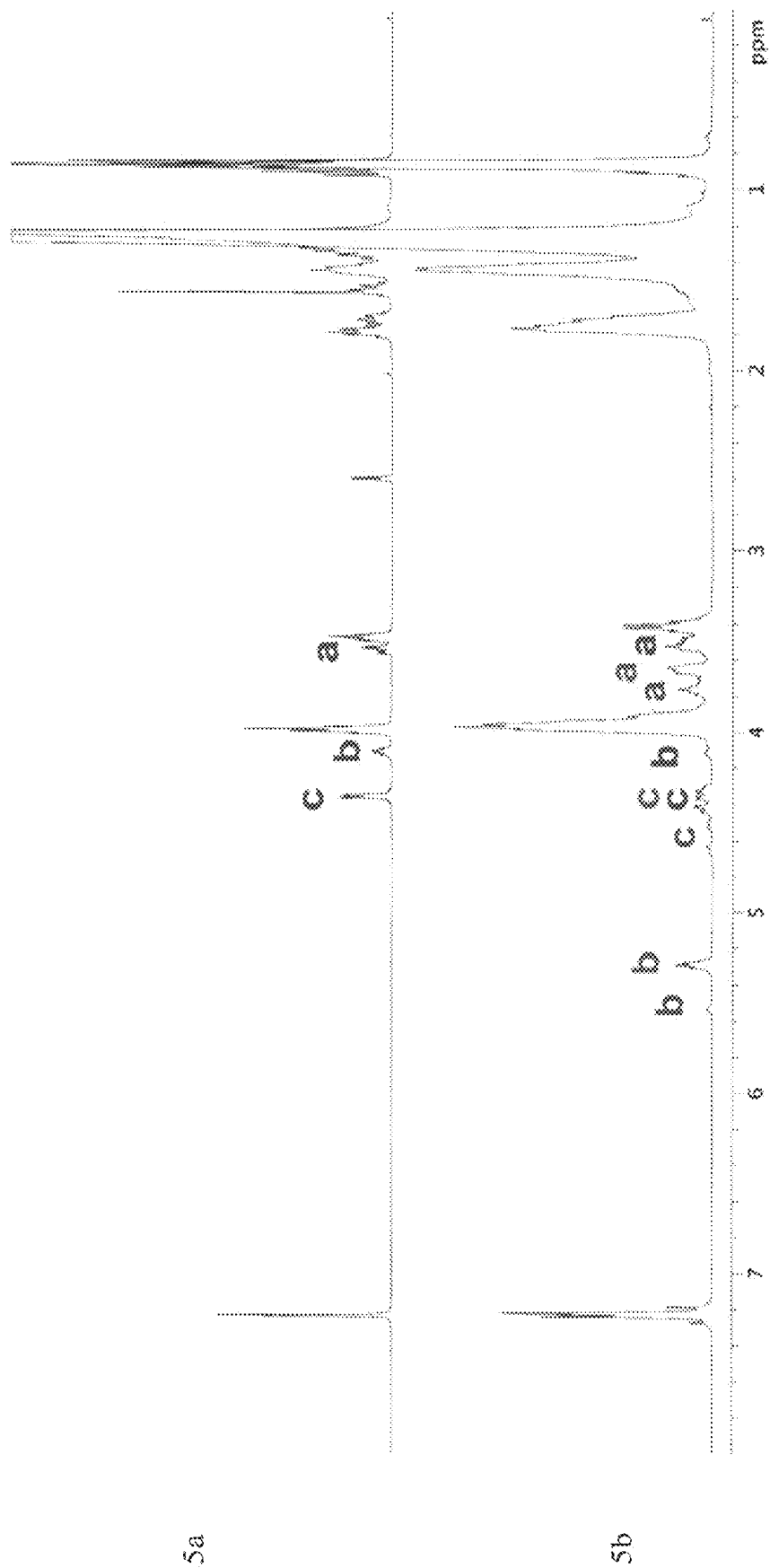


Fig. 5B

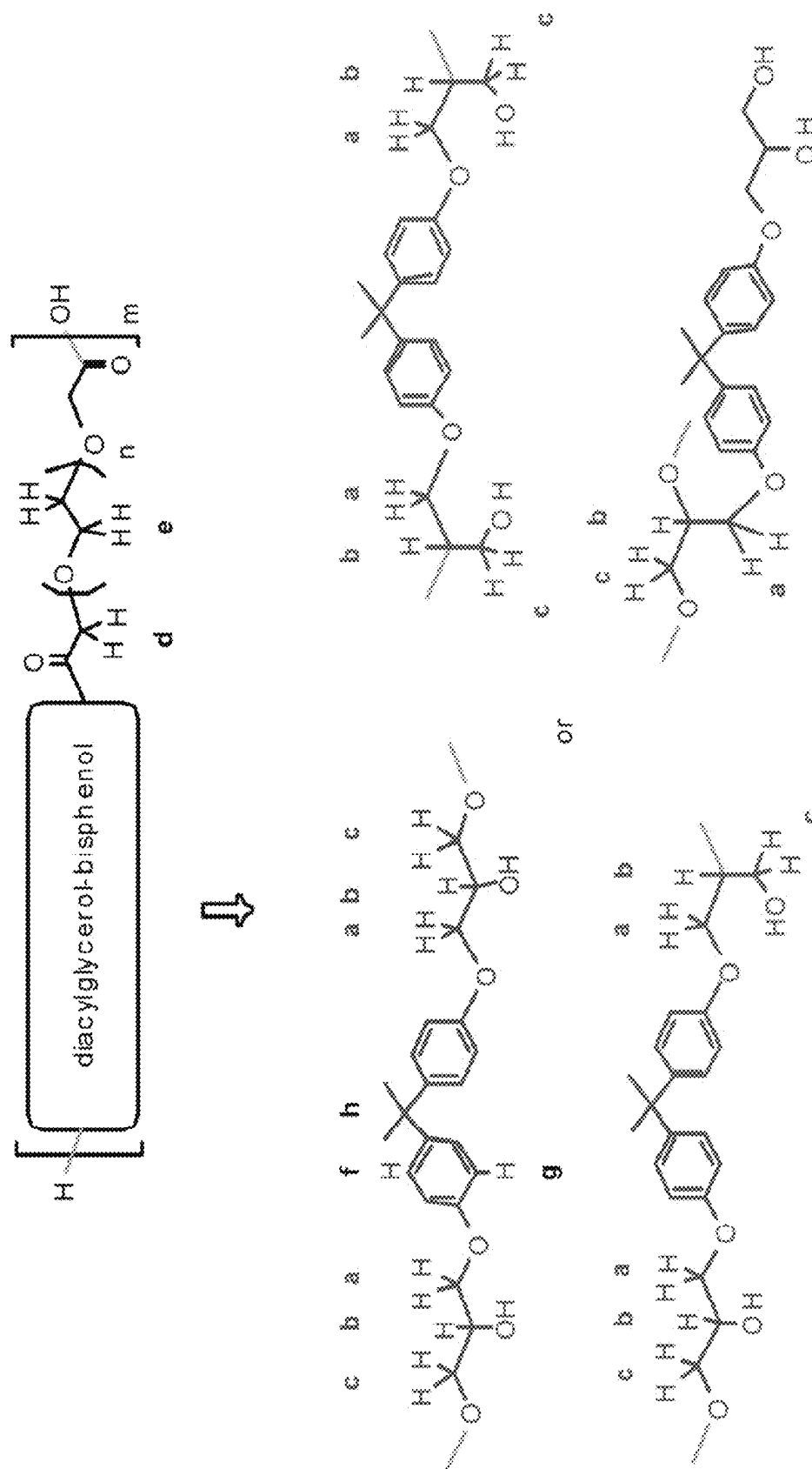


Fig. 6A

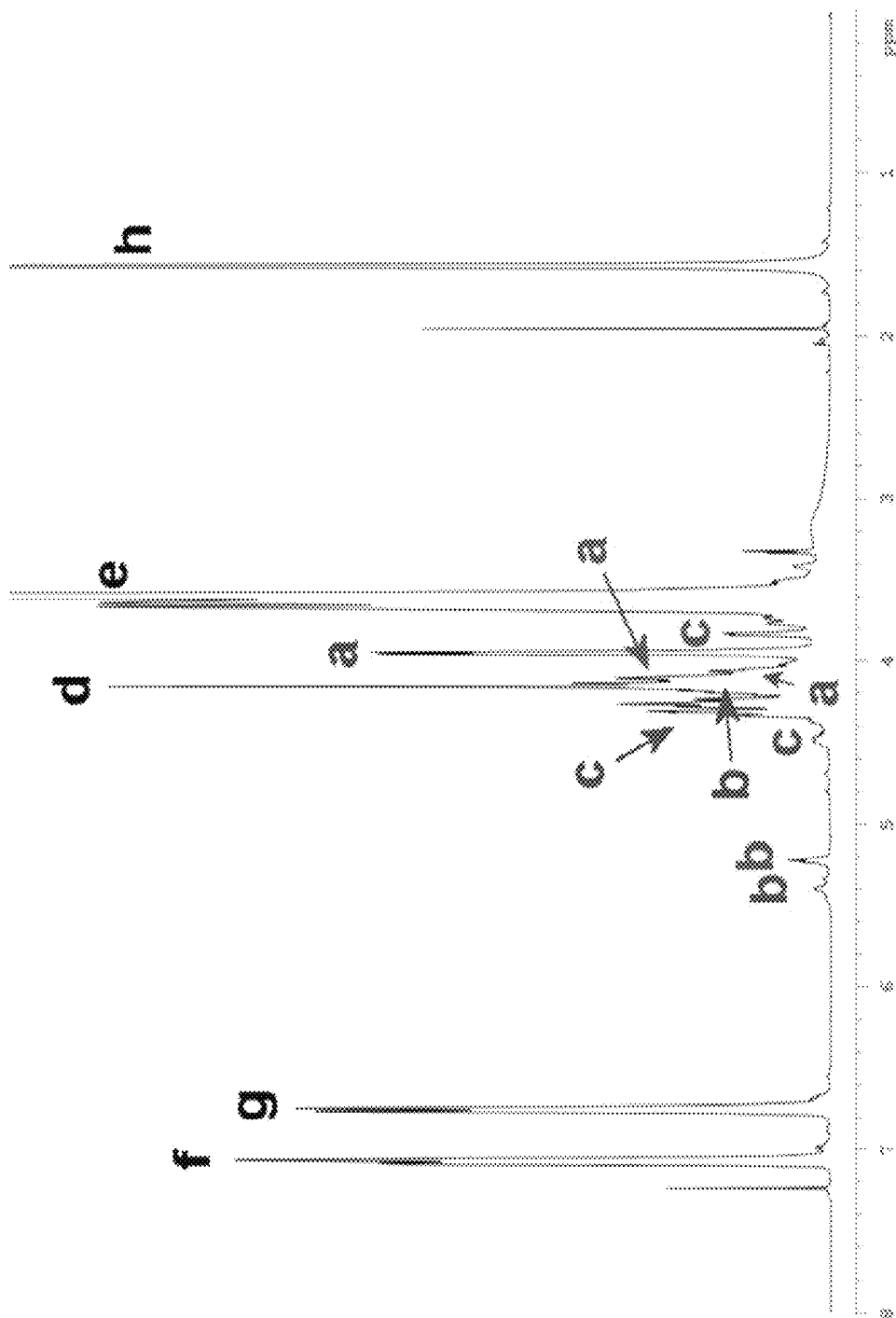


Fig. 6B

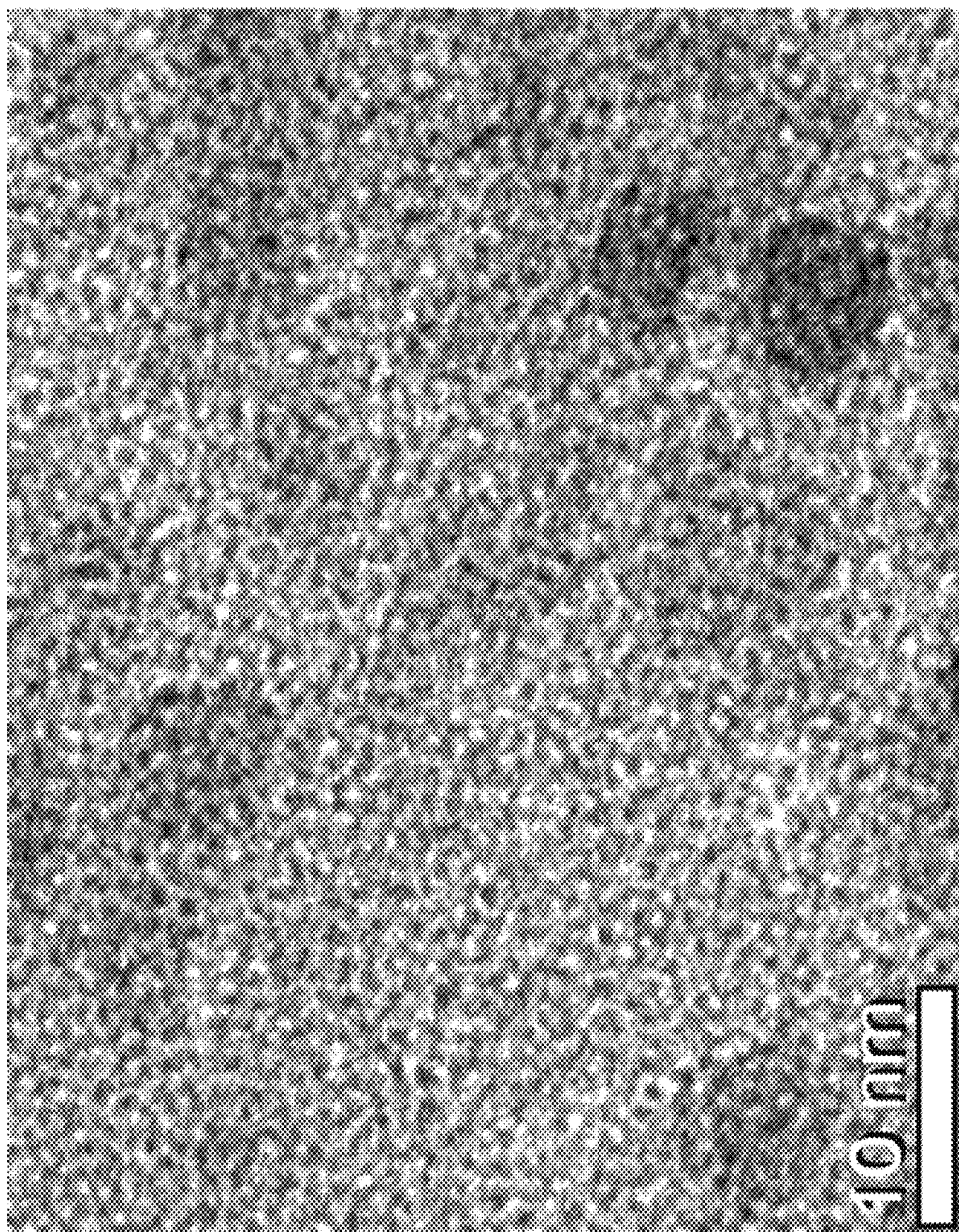


Fig. 7A

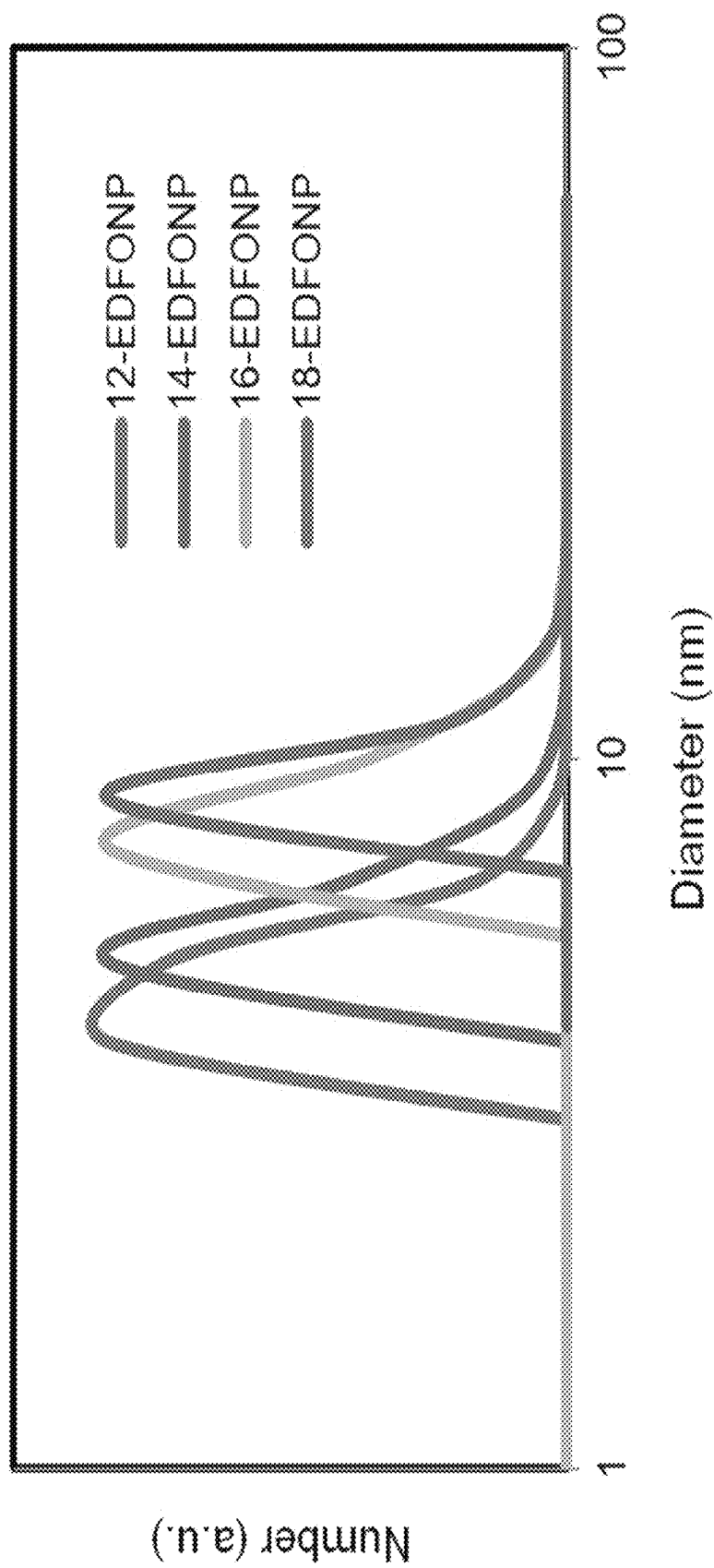


Fig. 7B

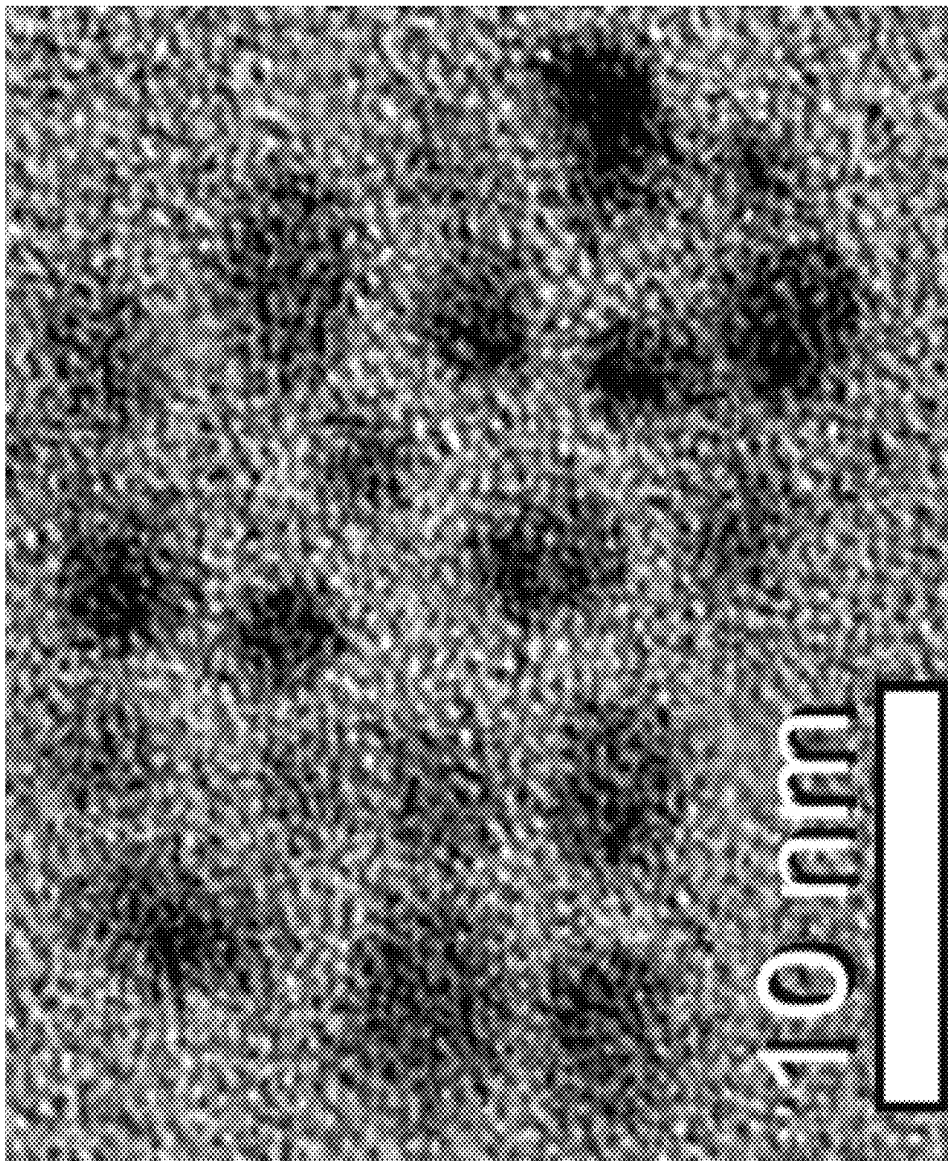


Fig. 7C

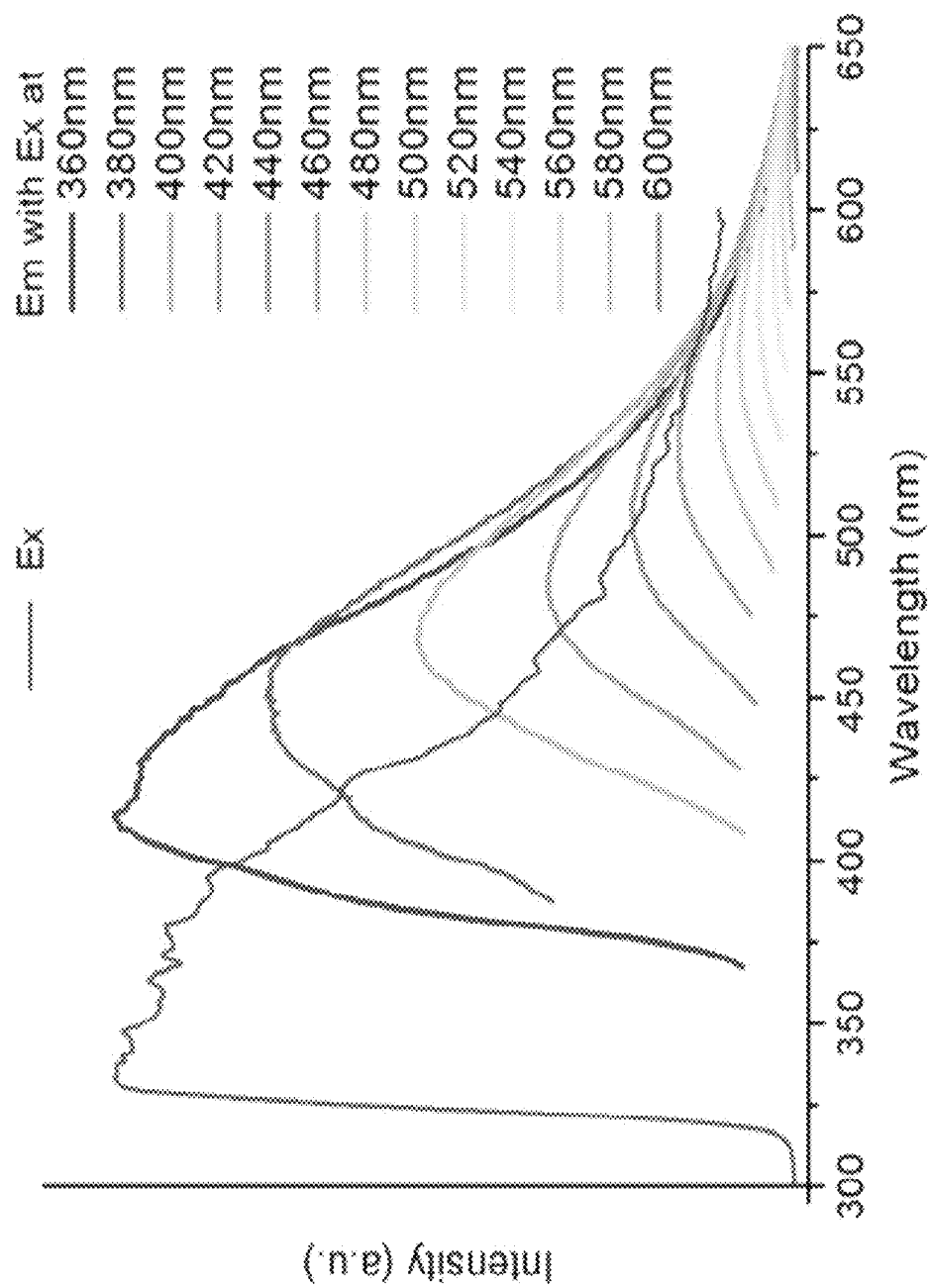


Fig. 8A

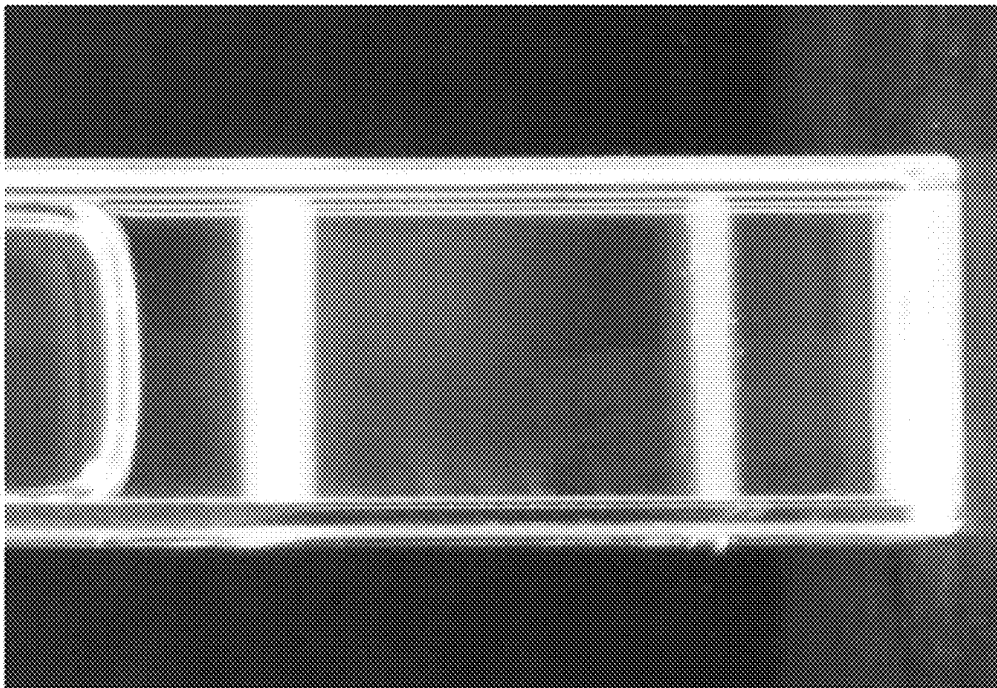


Fig. 8B

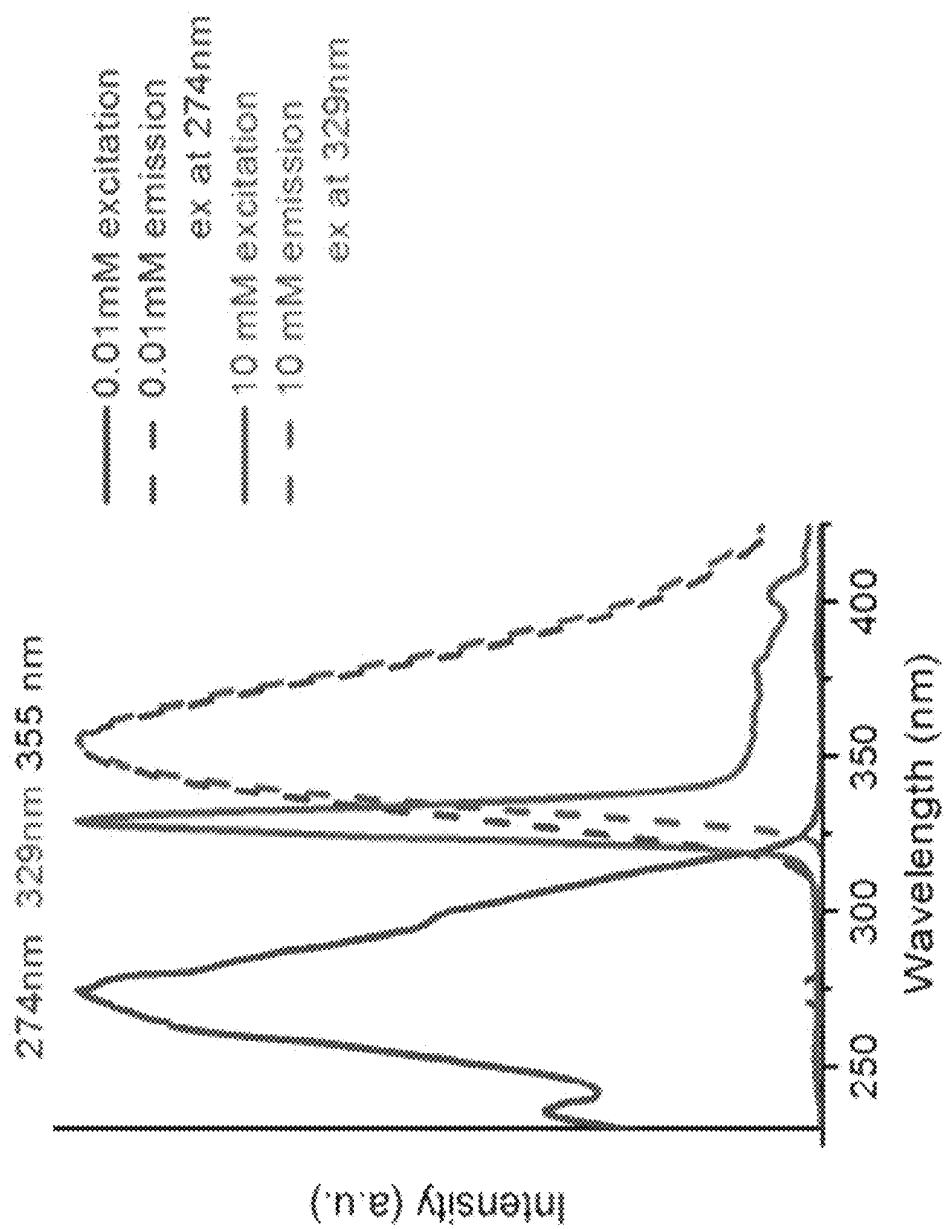


Fig. 9A

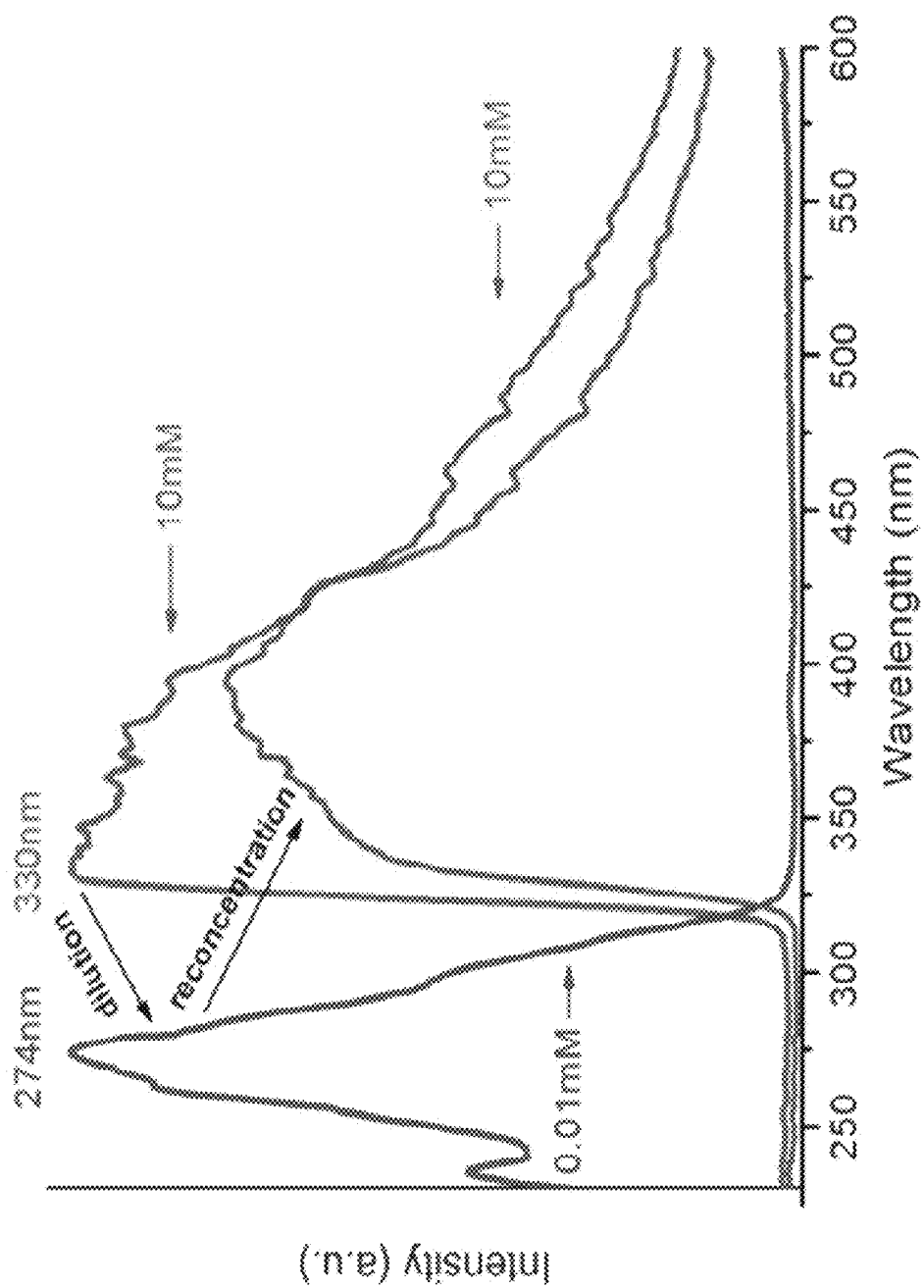


Fig. 9B

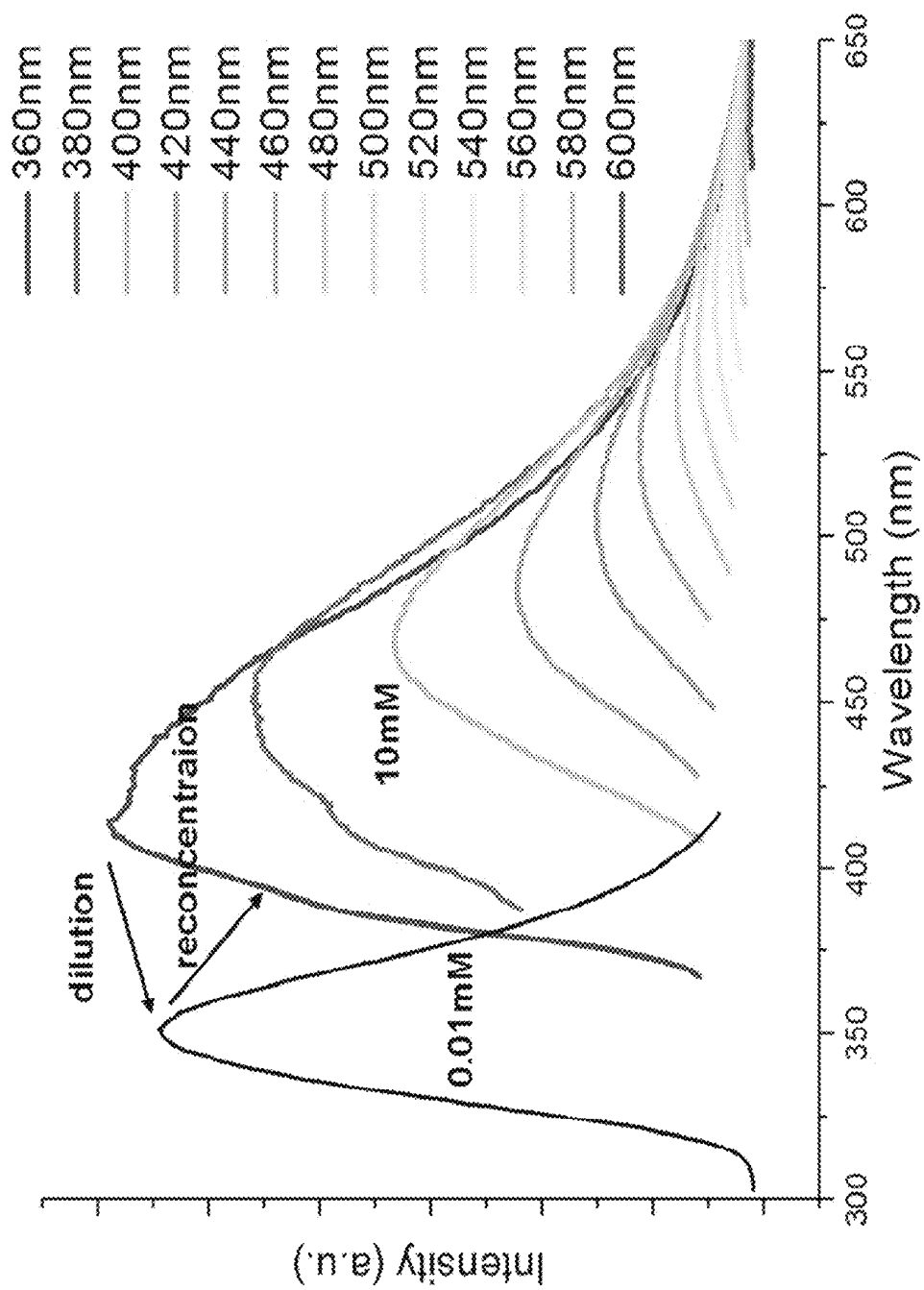


Fig. 9C

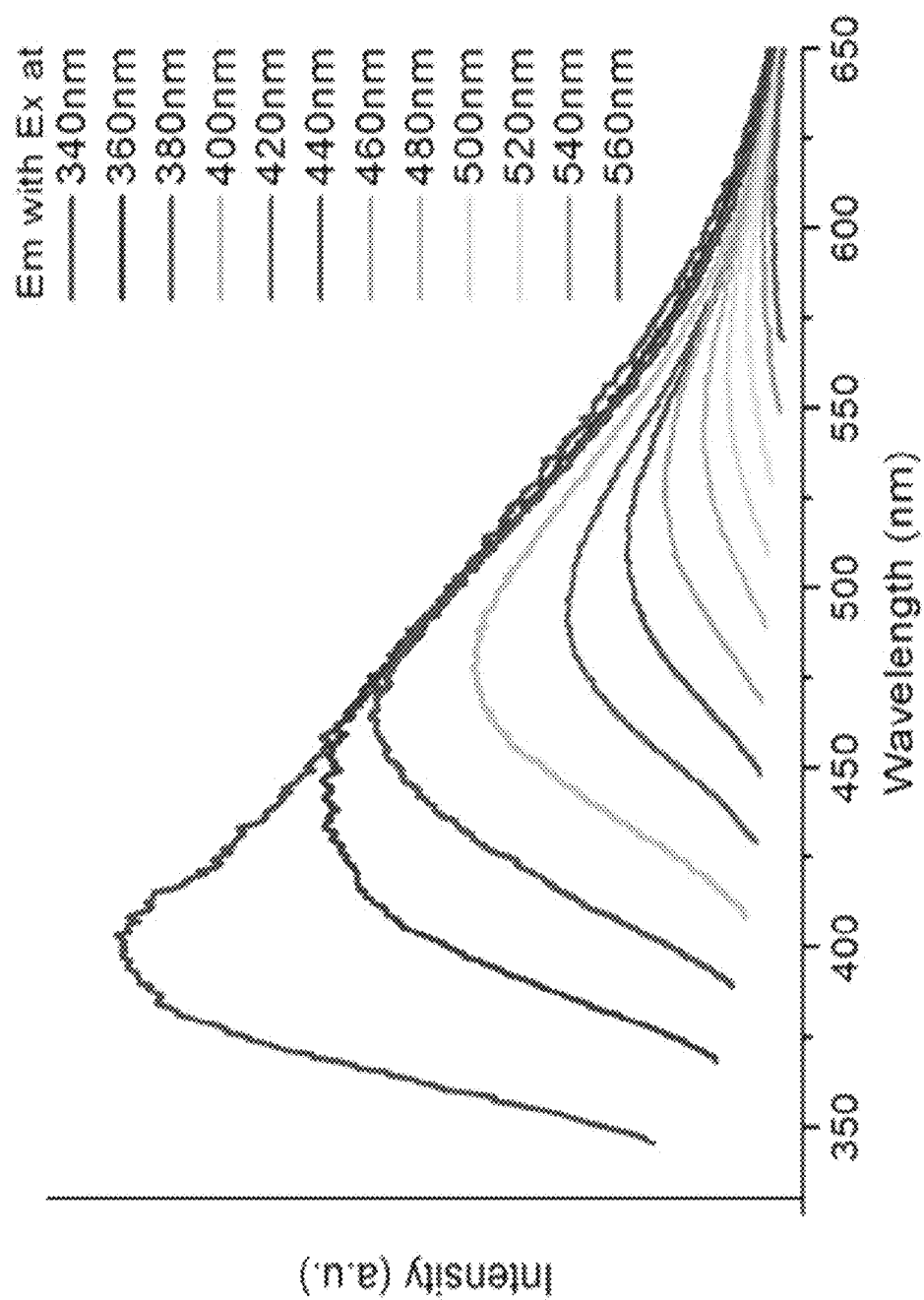


Fig. 10

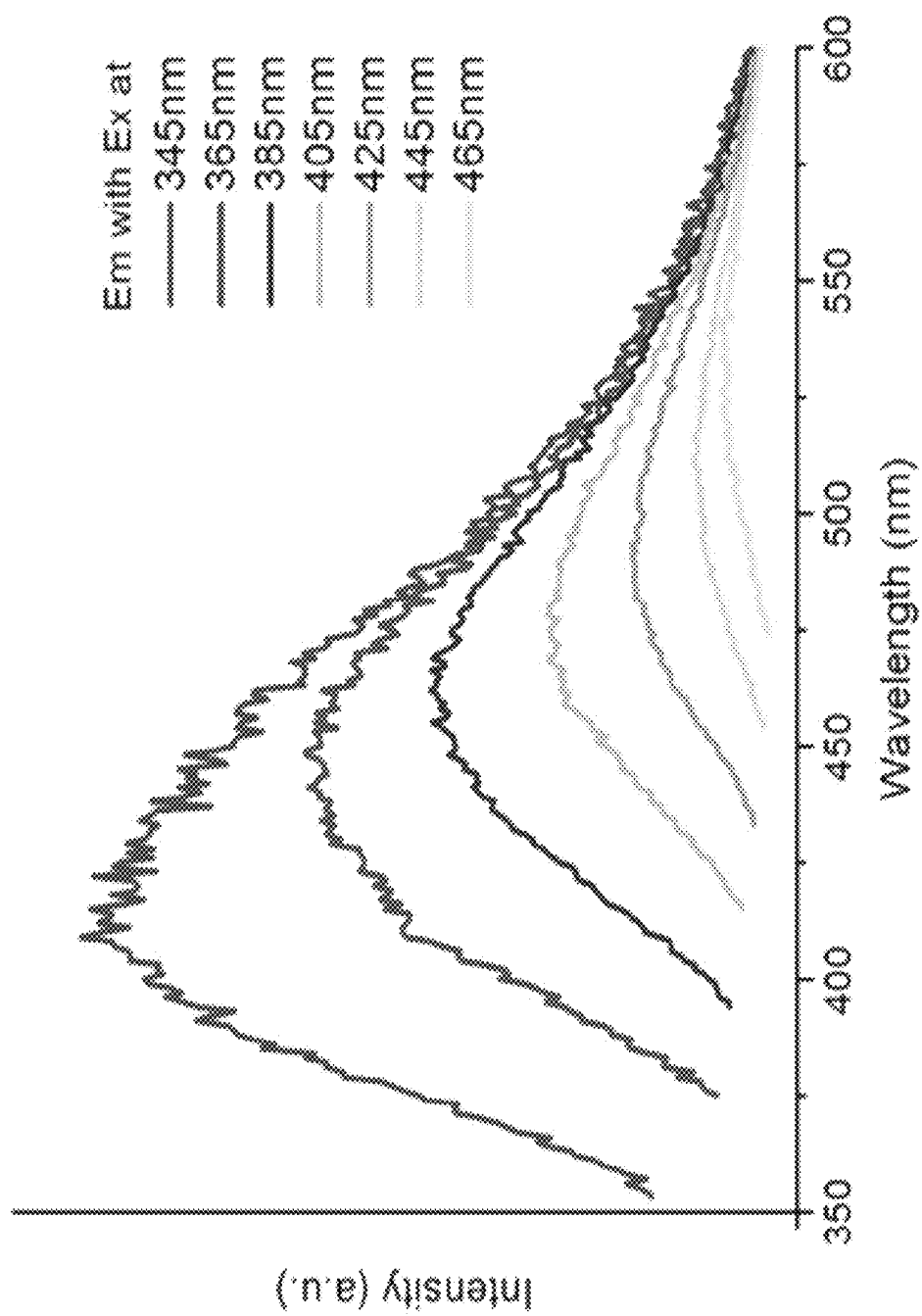


Fig. 11

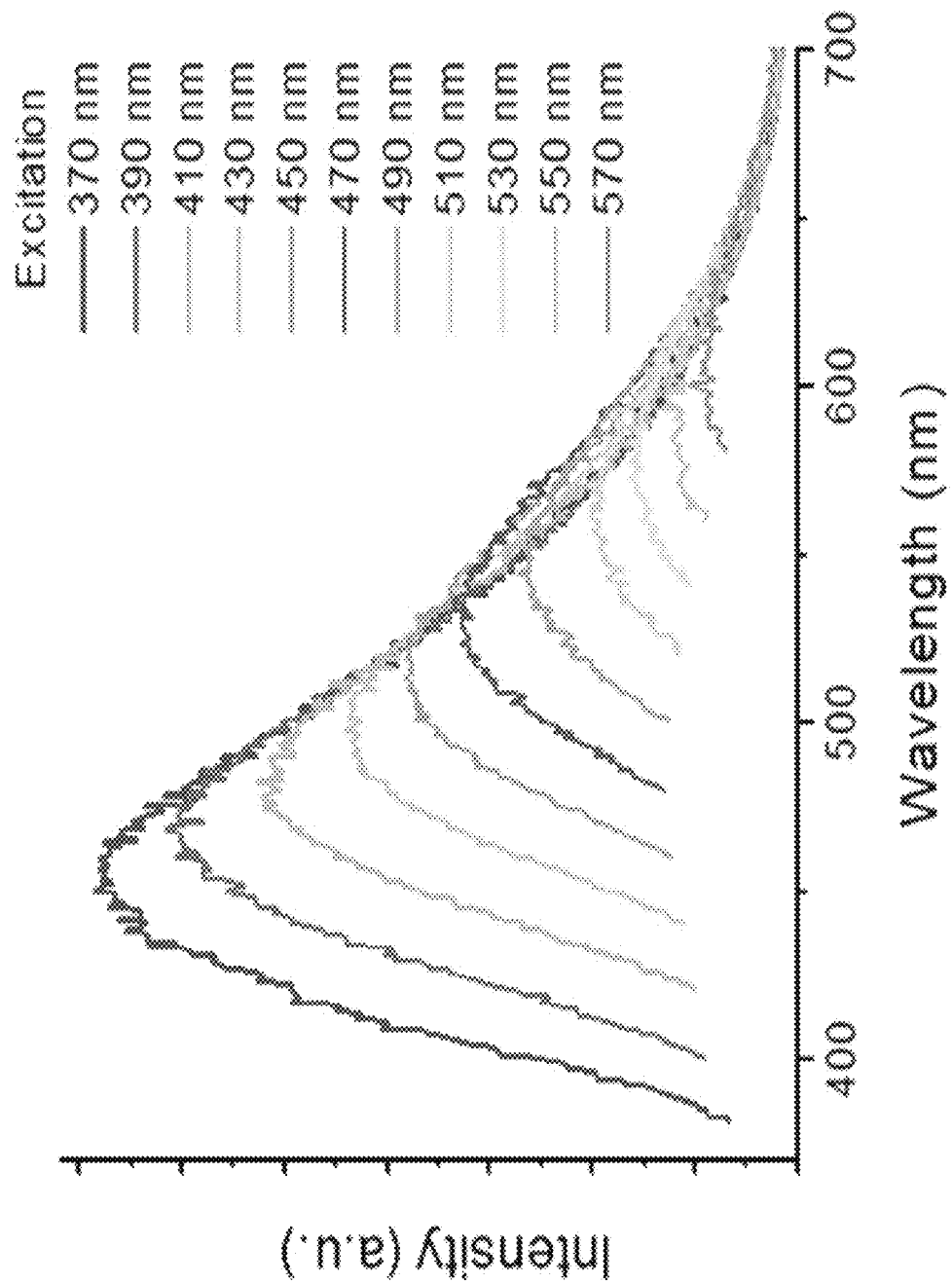


Fig. 12

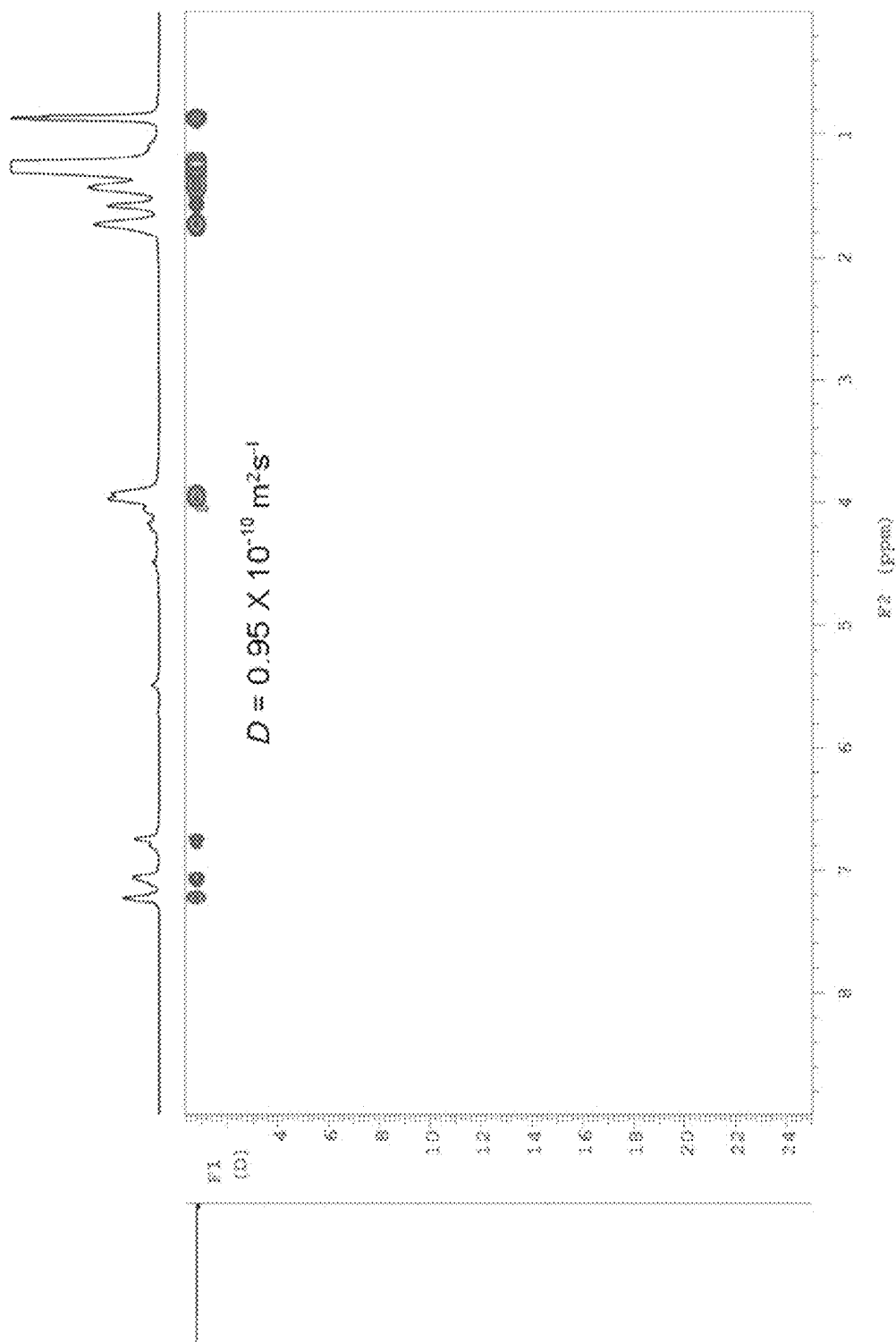


Fig. 13

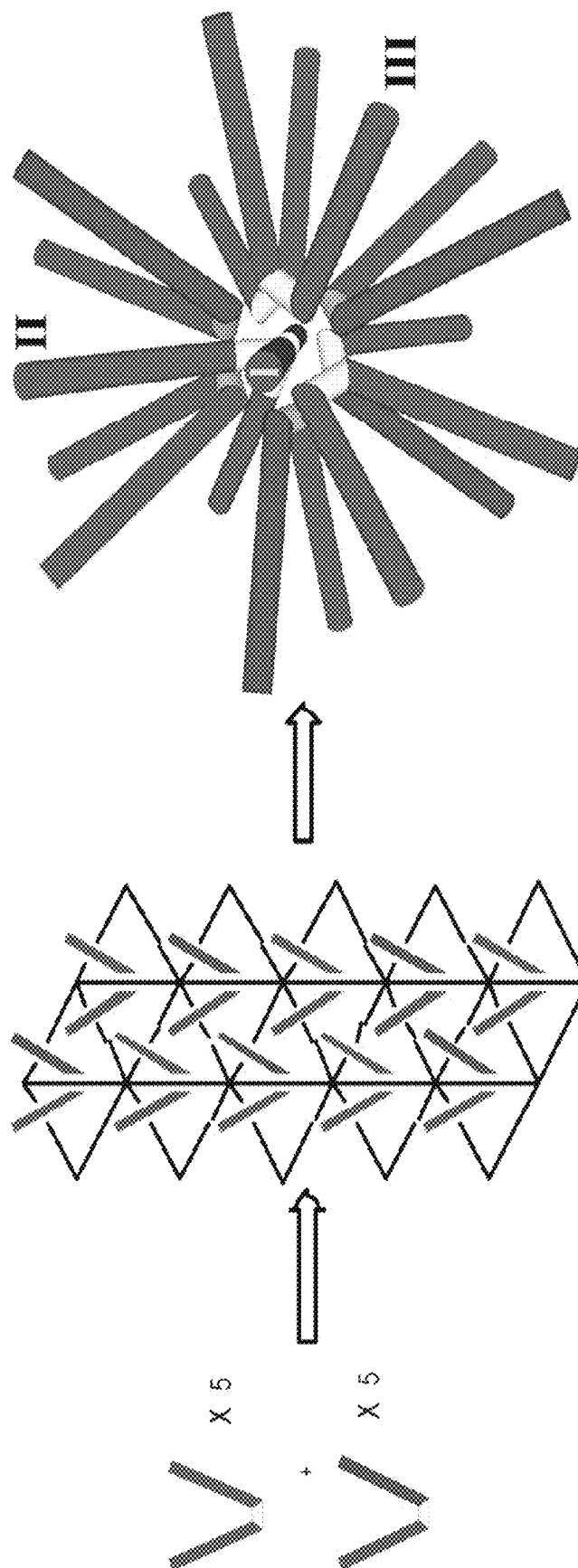


Fig. 14

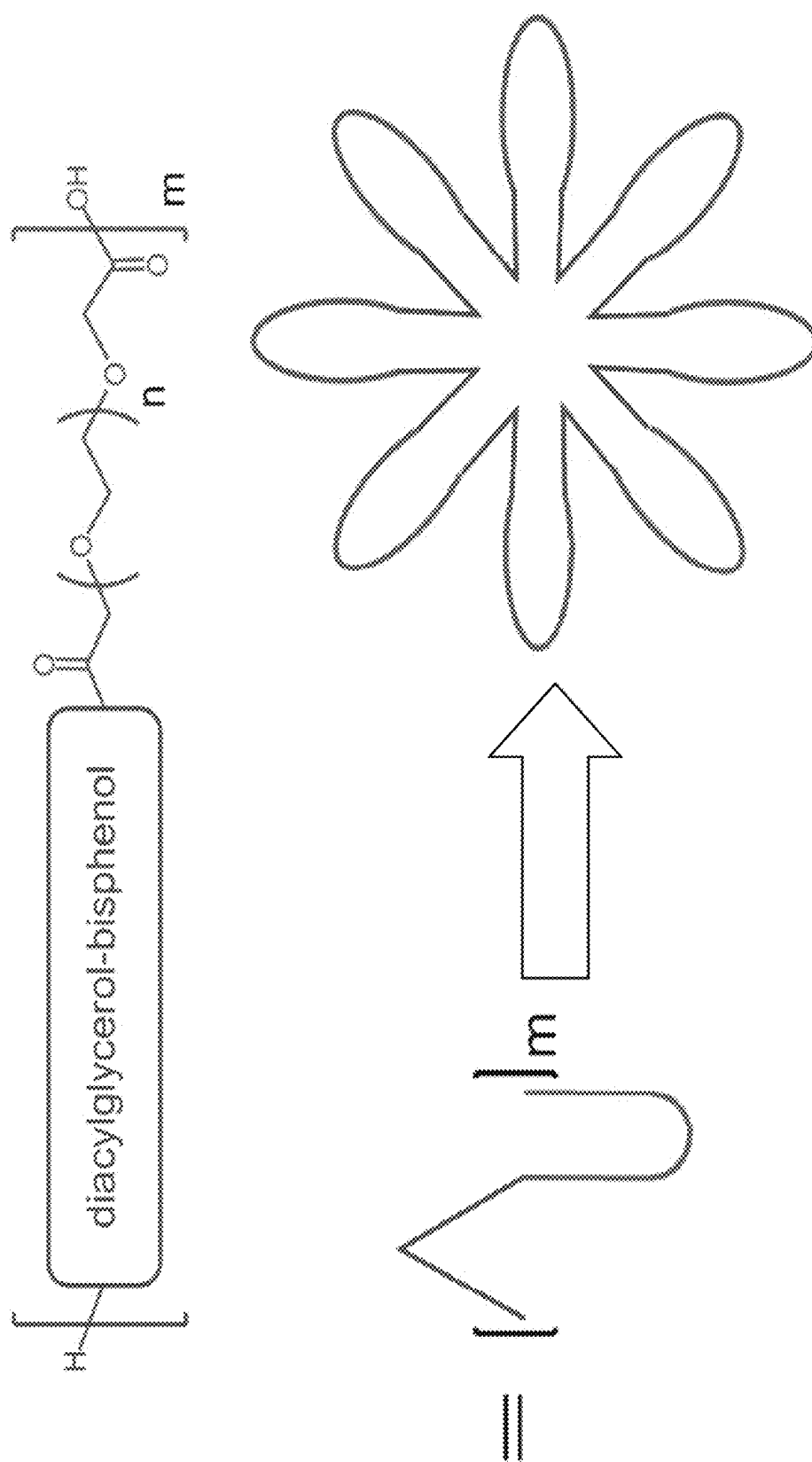


Fig. 15

1

# EXCITATION-DEPENDENT FLUORESCENT ORGANIC NANOPARTICLES

## BACKGROUND

### Technical Field

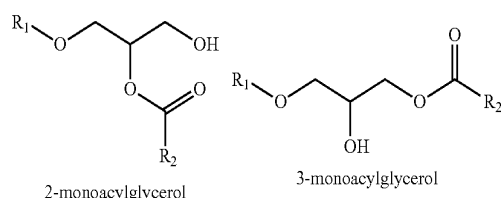
The disclosure relates to a fluorescent material. More particularly, the disclosure relates to a fluorescent organic nanoparticle.

### Description of Related Art

Organic fluorescent nanoparticles, excitation-dependent photoluminescence (or excitation-dependent fluorescent; EDF), hydrogenbonded clusters and lysobisphosphatidic acid are four interesting individual topics in materials and biological sciences. They have attracted much attention not only because of their unique properties and important applications, but also because the nature of their intriguing phenomena remained unclear.

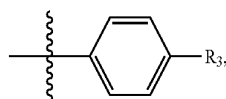
## SUMMARY

In one aspect, the present invention is directed to an excitation-dependent fluorescent organic nanoparticles. Each of the excitation-dependent fluorescent organic nanoparticles comprises a cluster of aggregated molecules of 2-monoacylglycerol and 3-monoacylglycerol having the chemical structures shown below:



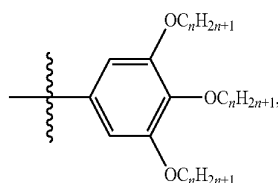
wherein the 2-monoacylglycerol is the major component. According to an embodiment,  $R_1$  is an aliphatic group with no more than 30 carbons or an aromatic group with no more than 30 carbons, and  $R_2$  is an aliphatic group with no more than 30 carbons or an aromatic group with no more than 30 carbons.

According to another embodiment, the  $R_1$  comprises



and  $R_3$  is an aliphatic group or an aromatic group.

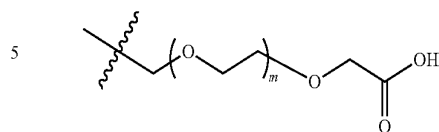
According to yet another embodiment, the  $R_2$  comprises



and  $n=10-20$ .

2

According to yet another embodiment,  $R_2$  comprises



and  $m$  is 10-14.

In another aspect, this invention directs to a method of forming an excitation-dependent fluorescent nanoparticles. The method comprises performing a ring-opening reaction and a radical catalyzed 3,2-acyl migration reaction. The ring-opening reaction is performed by reacting an epoxy glycidyl ether and an carboxylic acid to form a first mixture of the 3-monoacylglycerol and the 2-monoacylglycerol, wherein the 3-monoacylglycerol is the major component. The radical catalyzed 3,2-acyl migration reaction is performed by adding a catalyst to the first mixture of the 3-monoacylglycerol and the 2-monoacylglycerol to form the excitation-dependent fluorescent nanoparticles above.

The foregoing presents a simplified summary of the disclosure in order to provide a basic understanding to the reader. This summary is not an extensive overview of the disclosure and it does not identify key/critical elements of the present invention or delineate the scope of the present invention. Its sole purpose is to present some concepts disclosed herein in a simplified form as a prelude to the more detailed description that is presented later. Many of the attendant features will be more readily appreciated as the same becomes better understood by reference to the following detailed description considered in connection with the accompanying drawings.

## BRIEF DESCRIPTION OF THE DRAWINGS

The patent or application file contains at least one drawing executed in color. Copies of this patent or patent application publication with color drawing(s) will be provided by the Office upon request and payment of the necessary fee.

FIG. 1A is a diagram of the chemical structure of 2,2'-12-BTBA and the 3,3'-12-BTBA, and the marks for H and C atoms are also shown.

FIGS. 1B and 1C are the  $^1\text{H}$  and DEPT-135 NMR spectra of 12-No-EDF, respectively.

FIGS. 2A and 2B are the  $^1\text{H}$  and DEPT-135 NMR spectra of 12-EDFONP, respectively.

FIG. 2C shows the chemical structures of 2,2'-12-BTBA, 2,3-12-BTBA, 3,3'-12-BTBA, and 2,3-diacylglycerol bisphenol A.

FIG. 2D is a part of two-dimensional  $^1\text{H}$ - $^{13}\text{C}$  HSQC spectrum of 12-EDFONP in  $\text{CDCl}_3$  at room temperature.

FIG. 3A shows the chemical structures of 2,2'-12-BTBA, 2,3-12-BTBA, 3,3'-12-BTBA and free 2,3-12-diacylglycerol-bisphenol-glycerol isomer.

FIG. 3B is the  $^1\text{H}$  NMR spectra of 12-No-EDF (spectrum 3a), product obtained by treating 12-No-EDF with  $(\text{NH}_4)_2\text{S}_2\text{O}_8$  (spectrum 3b), and product obtained by treating 12-No-EDF with  $\text{K}_2\text{CO}_3$  (spectrum 3c).

FIG. 4A shows the chemical structures of 2,2'-17-BTBA, 2,3-17-BTBA, 3,3'-17-BTBA and 2,3-17-diacylglycerol-bisphenol-glycerol.

FIG. 4B is the  $^1\text{H}$  NMR spectra of 17-No-EDF (spectrum 4a) and 17-EDFONP (spectrum 4b).

FIG. 5A shows the chemical structures of the components of the 16-BGE-EDFONP.

3

FIG. 5B is the  $^1\text{H}$  NMR spectra of 16-BGE-No-EDF (spectrum 5a) and 16-BGE-EDFONP (spectrum 5b).

FIG. 6A shows the chemical structures of the EDF-polymer.

FIG. 6B is the  $^1\text{H}$  NMR spectrum of the EDF-polymer in  $\text{CDCl}_3$  at room temperature.

FIG. 7A is the high resolution transmission electron microscopy (HRTEM) image of 18-EDFONP.

FIG. 7B is a diagram showing mean diameters, measured by dynamic light scattering, of 12-EDFONP to 18-EDFONP in  $\text{CH}_2\text{Cl}_2$ .

FIG. 7C is a HRTEM image of the EDF-polymer.

FIG. 8A is excitation and emission spectra of 12-EDFONP at a concentration of 10 mM with a excitation-dependent visible fluorescence from 410-620 nm.

FIG. 8B is a fluorescent photograph of 12-EDFONP in a  $\text{CH}_2\text{Cl}_2$  solution.

FIG. 9A is excitation and emission spectra of 12-No-EDF at concentrations of 0.01 and 10 mM.

FIGS. 9B and 9C are excitation and emission spectra of 12-EDFONP at an initial concentration of 10 mM (red line), then diluted to 0.01 mM (blue line) and reconcentrated back to 10 mM (green line), respectively.

FIG. 10 is emission spectrum of 17-EDFONP based on stearic acid at a concentration of 10 mM with a visible EDF phenomena.

FIG. 11 is emission spectrum of 16-BGE-EDFONP at a concentration of 10 mM with a visible EDF phenomena.

FIG. 12 is emission spectrum of the EDF-Polymer.

FIG. 13 is DOSY  $^1\text{H}$  NMR 2D spectrum of 18-EDFONP in a solution.

FIG. 14 is a diagram showing the formation of Great Stellated Dodecahedral decamers by the self-assembly of five 2,2'-n-BTBA and five 2,3-n-BTBA.

FIG. 15 is a cartoon model of nanoparticle with bisphenol A aggregates and poly(ethylene glycol) moieties serving as core and shell compartments respectively.

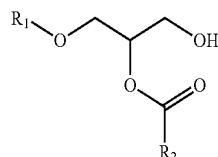
### DETAILED DESCRIPTION

In the following detailed description, for purposes of explanation, numerous specific details are set forth in order to provide a thorough understanding of the disclosed embodiments. It will be apparent, however, that one or more embodiments may be practiced without these specific details. In other instances, well-known structures and devices are schematically shown in order to simplify the drawing.

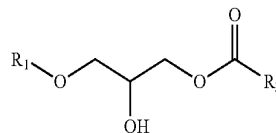
#### Excitation-Dependent Fluorescent Organic Nanoparticles

The composition of excitation-dependent fluorescent organic nanoparticles (abbreviated as EDFONPs) comprises at least a cluster of aggregated molecules of 2-monoacylglycerol and 3-monoacylglycerol having the chemical structures shown below:

4

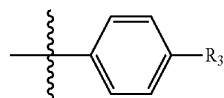


2-monoacylglycerol  
Major component

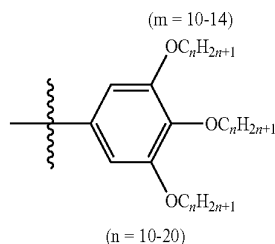
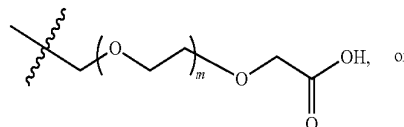


3-monoacylglycerol  
Minor component

$R_1$  above can be an aliphatic group with no more than 30 carbons or an aromatic group with no more than 30 carbon, such as an alkyl chain, or



and  $R_3$  is an aliphatic group with no more than 30 carbons or an aromatic group with no more than 30 carbons.  $R_2$  above can be an aliphatic group or an aromatic group, such as an alkyl chain,

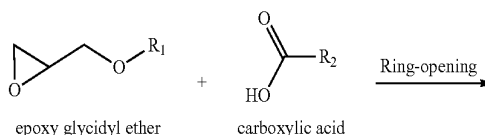


In this cluster, the 2-monoacylglycerol is the major component.

#### Preparation of Excitation-Dependent Fluorescent Organic Nanoparticles

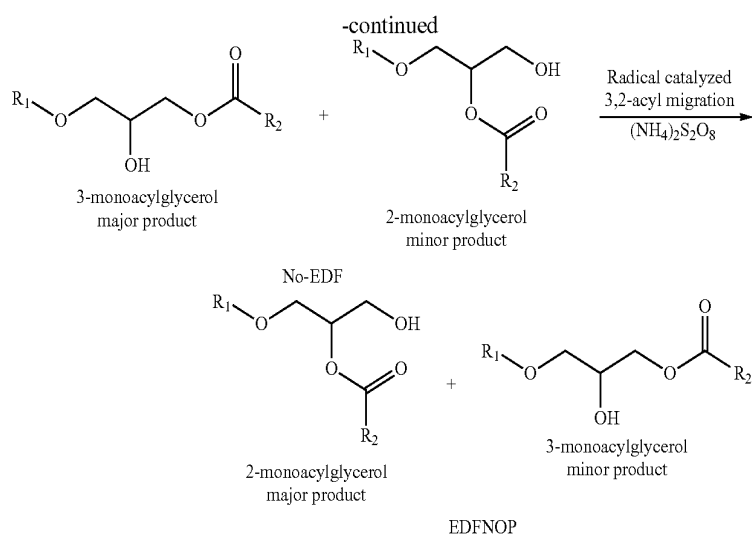
EDFONPs are prepared by two sequential reactions. The first reaction is a ring-opening reaction, and the second reaction is a radical-catalyzed 3,2-acyl migration reaction. The reaction formula is shown in Scheme 1.

Scheme 1



5

6



In scheme 1, the  $R_1$  of epoxy glycidyl ether can be an aliphatic group or an aromatic group, and the  $R_2$  of the carboxylic acid can be an aliphatic group or an aromatic group, as described above. For example, the  $R_1$  of the epoxy glycidyl ether can be butyl, octyl, decyl, benzyl, or bisphenol A groups, and the carboxylic acid can be dodecanoic acid, heptadecanoic acid, 3,4,5-tris(n-alkoxyl)benzoic acid, or polyethylene glycol diacid.

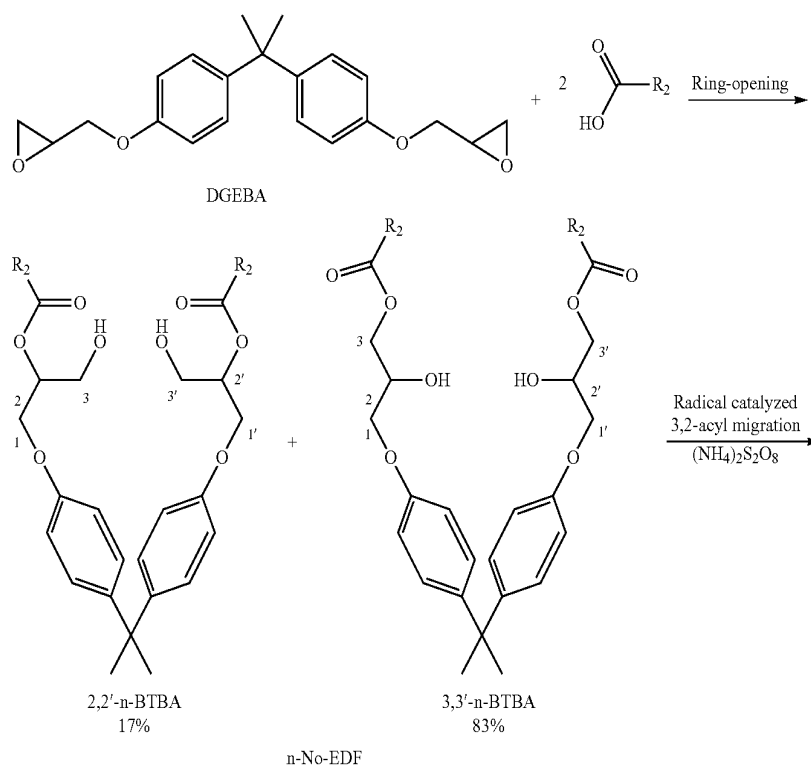
In the first reaction, the initial products containing a mixture of a 2-positional isomer as the minor component and a 3-positional isomer as the major component are obtained via a ring-opening reaction by treating the glycidyl ether epoxy with the carboxylic acid. The initial products are abbreviated as No-EDF because no EDF behavior was observed.

In the second reaction, the radical catalyzed 3,2-acyl migration reaction is conducted by treating the initial products with ammonium persulfate  $((NH_4)_2S_2O_8)$  as a catalyst under a neat condition. The second products resulting from the migration of the 3,2-acyl contains a mixture of the 2-positional isomer as the major component and the 3-positional isomer as the minor component. The second products are abbreviated as EDFNOP because EDF behavior was observed.

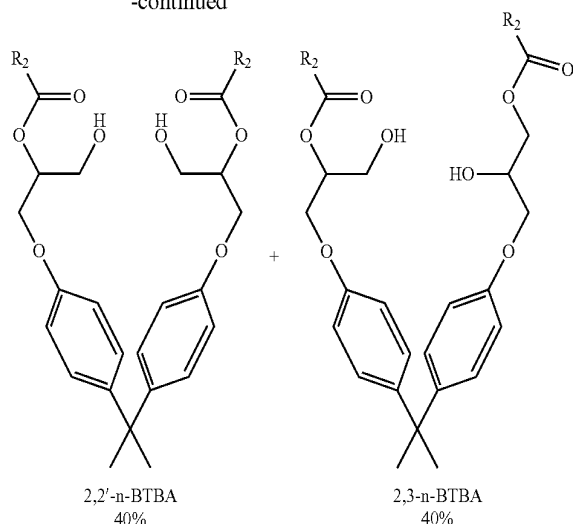
Some examples of the above reactions in scheme 1 are provided below.

Embodiment 1: Synthesis of n-No-EDF and n-EDFNOP

Scheme 2

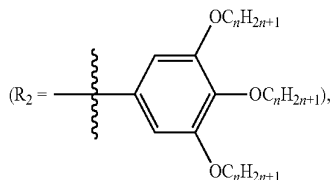


-continued



n-EDFONP

In Scheme 2 of this embodiment, the epoxy glycidyl ether in scheme 1 was diglycidyl ether of bisphenol A (DGEBA), and the carboxylic acid ( $R_2$ -COOH) was 3,4,5-tris(n-alkoxy)benzoic acid



and the carbon number of the alkoxy group, n, was 12, 14, 16, and 18, respectively.

The obtained initial products No-EDF in Scheme 2 of the first ring opening reaction are denoted as n-No-EDF, and n=12, 14, 16, and 18, respectively. The initial products were a mixture of 2,2'-n-bis(3,4,5-tris(n-alkoxy)monobenzoylglyceride)bisphenol A (abbreviated as 2,2'-n-BTBA) and the 3,3'-n-BTBA, which were identified by  $^1\text{H}$ ,  $^{13}\text{C}$ , heteronuclear single quantum correlation (HSQC) and heteronuclear multiple bond correlation (HMBC) NMR experiments.

The obtained final products EDFONP in Scheme 2 of the second 3,2-acyl migration reaction are denoted as n-EDFONP, and n=12, 14, 16, and 18, respectively. Accordingly, the final products were a mixture of 2,2'-n-BTBA and the 2,3-n-BTBA, and also identified through  $^1\text{H}$ ,  $^{13}\text{C}$ , HSQC and HMBC NMR spectra, but their NMR spectra are significantly different from that of the n-No-EDFs.

The detailed synthesis of 12-No-EDF and 12-EDFONP are described below to be a representative example, because the same procedure was systematically used for all the members of the series.

**Synthesis of 12-No-EDF:** A solution of bisphenol A diglycidyl ether (0.68 g, 2.00 mmol), 3,4,5-tris(dodecyloxy) benzoic acid (2.80 g, 4.15 mmol) and tetra-n-butylammonium bromide (TBAB) (0.32 g, 1.00 mmol) in mix-solvent of acetonitrile (40 ml) and xylene (20 ml) was reflux at  $85^\circ\text{C}$  for 24 h. The solvent was removed under reduced pressure. The crude product was recrystallized twice from ethanol, and a white solid is obtained (2.30 g, 68%). The

crude product was composed of 2,2'-12-BTBA and the 3,3'-12-BTBA positional isomers.

FIG. 1A is a diagram of the chemical structure of 2,2'-12-BTBA and the 3,3'-12-BTBA, and the marks for H and C atoms are also shown. FIGS. 1B and 1C are the  $^1\text{H}$  and DEPT-135 NMR spectra of 12-No-EDF, respectively. From FIGS. 1B and 1C, it can be seen that the molar ratio of the 2,2'-12-BTBA (red e, d and f) and 3,3'-12-BTBA (blue e, d and f) isomers is approximately 17:83. The molar ratio is typical for kinetically controlled reactions because the 3-position of DGEBA contains less steric hindrance than the 2-position.

12-No-EDF:  $^1\text{H}$  NMR (600 MHz,  $\text{CDCl}_3$ ):  $\delta$  7.26 (s, 4H), 7.14 (d, 4H,  $J=7.8$  Hz), 6.83 (d, 4H,  $J=7.8$  Hz), 5.41 (m, 0.34H, 2,2'-12-BTBA), 4.51 (m, 3.32H, 3,3'-12-BTBA), 4.34 (m, 1.66 H, 3,3'-12-BTBA), 4.26 (m, 0.68H, 2,2'-12-BTBA), 4.08 (m, 3.32H, 3,3'-12-BTBA), 4.02 (m, 12.68H), 2.75 (b, 2H), 1.78 (m, 12H), 1.63 (s, 6H), 1.47 (m, 12H), 1.34 (m, 96H), 0.88 (t, 18H,  $J=6.6$  Hz);  $^{13}\text{C}$  NMR (150 MHz,  $\text{CDCl}_3$ ):  $\delta$  166.8, 156.2, 152.9, 143.8, 142.8, 127.6, 124.1, 114.0, 108.4, 77.1, 73.5, 69.3, 68.9 (3,3'-12-BTBA), 68.8 (3,3'-12-BTBA), 66.4 (2,2'-12-BTBA), 66.1 (3,3'-12-BTBA), 62.3 (2,2'-12-BTBA), 41.8, 31.9-22.7, 14.1; Elementary analysis (calcd., found for  $\text{C}_{107}\text{H}_{180}\text{O}_{14}$ ): C (76.02, 76.01), H (10.73, 10.79).

14-No-EDF:  $^1\text{H}$  NMR (600 MHz,  $\text{CDCl}_3$ ):  $\delta$  7.26 (s, 4H), 7.14 (d, 4H,  $J=7.8$  Hz), 6.83 (d, 4H,  $J=7.8$  Hz), 5.41 (m, 0.34H, 2,2'-14-BTBA), 4.51 (m, 3.32H, 3,3'-14-BTBA), 4.34 (m, 1.66 H, 3,3'-14-BTBA), 4.26 (m, 0.68H, 2,2'-14-BTBA), 4.08 (m, 3.32H, 3,3'-14-BTBA), 4.02 (m, 12.68H), 2.75 (b, 2H), 1.78 (m, 12H), 1.63 (s, 6H), 1.47 (m, 12H), 1.34 (m, 120H), 0.88 (t, 18H,  $J=6.6$  Hz);  $^{13}\text{C}$  NMR (150 MHz,  $\text{CDCl}_3$ ):  $\delta$  166.8, 156.2, 152.9, 143.8, 142.8, 127.6, 124.1, 114.0, 108.4, 77.1, 73.5, 69.3, 68.9 (3,3'-14-BTBA), 68.8 (3,3'-14-BTBA), 66.4 (2,2'-14-BTBA), 66.1 (3,3'-14-BTBA), 62.3 (2,2'-14-BTBA), 41.8, 31.9-22.7, 14.1; Elementary analysis (calcd., found for  $\text{C}_{119}\text{H}_{204}\text{O}_{14}$ ): C (76.89, 76.87), H (11.06, 11.03).

16-No-EDF:  $^1\text{H}$  NMR (600 MHz,  $\text{CDCl}_3$ ):  $\delta$  7.26 (s, 4H), 7.14 (d, 4H,  $J=7.8$  Hz), 6.83 (d, 4H,  $J=7.8$  Hz), 5.41 (m, 0.34H, 2,2'-16-BTBA), 4.51 (m, 3.32H, 3,3'-16-BTBA), 4.34 (m, 1.66 H, 3,3'-16-BTBA), 4.26 (m, 0.68H, 2,2'-16-BTBA), 4.08 (m, 3.32H, 3,3'-16-BTBA), 4.02 (m, 12.68H),

2.75 (b, 2H), 1.78 (m, 12H), 1.63 (s, 6H), 1.47 (m, 12H), 1.34 (m, 144H), 0.88 (t, 18H,  $J=6.6$  Hz);  $^{13}\text{C}$  NMR (150 MHz,  $\text{CDCl}_3$ ):  $\delta$  166.8, 156.2, 152.9, 143.8, 142.8, 127.6, 124.1, 114.0, 108.4, 77.1, 73.5, 69.3, 68.9 (3,3'-16-BTBA), 68.8 (3,3'-16-BTBA), 66.4 (2,2'-16-BTBA), 66.1 (3,3'-16-BTBA), 62.3 (2,2'-16-BTBA), 41.8, 31.9-22.7, 14.1; Elementary analysis (calcd., found for  $\text{C}_{131}\text{H}_{228}\text{O}_{14}$ ): C (77.61, 77.23), H (11.34, 11.28).

18-No-EDF:  $^1\text{H}$  NMR (600 MHz,  $\text{CDCl}_3$ ):  $\delta$  7.26 (s, 4H), 7.14 (d, 4H,  $J=7.8$  Hz), 6.83 (d, 4H,  $J=7.8$  Hz), 5.41 (m, 10 0.34H, 2,2'-18-BTBA), 4.51 (m, 3.32H, 3,3'-18-BTBA), 4.34 (m, 1.66 H, 3,3'-18-BTBA), 4.26 (m, 0.68H, 2,2'-18-BTBA), 4.08 (m, 3.32H, 3,3'-18-BTBA), 4.02 (m, 12.68H), 2.75 (b, 2H), 1.78 (m, 12H), 1.63 (s, 6H), 1.47 (m, 12H), 1.34 (m, 168H), 0.88 (t, 18H,  $J=6.6$  Hz);  $^{13}\text{C}$  NMR (150 MHz,  $\text{CDCl}_3$ ):  $\delta$  166.8, 156.2, 152.9, 143.8, 142.8, 127.6, 124.1, 114.0, 108.4, 77.1, 73.5, 69.3, 68.9 (3,3'-14-BTBA), 68.8 (3,3'-18-BTBA), 66.4 (2,2'-18-BTBA), 66.1 (3,3'-18-BTBA), 62.3 (2,2'-18-BTBA), 41.8, 31.9-22.7, 14.1; Elementary analysis (calcd., found for  $\text{C}_{143}\text{H}_{252}\text{O}_{14}$ ): C 20 (78.23, 77.94), H (11.57, 11.58).

Synthesis of 12-EDFONP: 12-No-EDF (0.7402 g, 0.44 mmol) and ammonium persulfate ( $(\text{NH}_4)_2\text{S}_2\text{O}_8$ ) (0.013 g, 0.057 mmol) were heated and stirred under  $\text{N}_2$  at  $150^\circ\text{C}$ . for 38 h in a neat condition. The crude product was then dissolved in  $\text{CH}_2\text{Cl}_2$  and filtered. The filtrate was dried under vacuum to give a brown product with a yield of 97% (0.72 g, 0.43 mmol). The brown product is composed of 2,2'-12-BTBA, 2,3-12-BTBA, 3,3'-12-BTBA and 2,3-12-diacylglycerol-bisphenol-glycerol isomers (a phosphatidylglycerol (PG)-like compound), and no non-acyl migrated side product is found.

FIGS. 2A and 2B are the  $^1\text{H}$  and DEPT-135 NMR spectra of 12-EDFONP, respectively. In FIG. 2A, the  $^1\text{H}$  signals of the 12-EDFONP in  $\text{CDCl}_3$  at 10 mM are composed of approximately 80% broad and 20% sharp signals. The dominant broad NMR signals are assigned to 2,2'-12-BTBA (40%) and 2,3-12-BTBA (40%), and they have significant upfield or downfield shifts compared with that of the initial 12-No-EDF. The remaining 20% sharp signals correspond to free 2,2'-12-BTBA (1%), free 3,3'-12-BTBA (15%) and free 2,3-12-diacylglycerol-bisphenol-glycerol isomer (4%), and the chemical structures thereof are shown in FIG. 2C.

These broad signals in FIG. 2A are attributed to the extremely dense aggregated structures in which the internal conformational mobility is reduced. In contrast, all of the  $^1\text{H}$  NMR signals corresponding to 2,2'-12-BTBA and 3,3'-12-BTBA in the 12-No-EDF in  $\text{CDCl}_3$  at 10 mM in FIG. 1A are normally sharp, which indicates no dense aggregated structure being formed in the low concentration of 2,2'-12-BTBA. In other words, the increased concentration of the 2-monoacylglycerol moieties in the 12-EDFONP from approximately 17% to 61% after the 3,2-acyl migration causes the formation of dense aggregated structures with a yield of 80%.

Remarkable differences in both the  $^{13}\text{C}$  and  $^1\text{H}$  chemical shifts are observed in the comparison of the 12-EDFONP and 12-No-EDF NMR spectra.

FIG. 2D is a part of two-dimensional  $^1\text{H}$ - $^{13}\text{C}$  HSQC spectrum of 12-EDFONP in  $\text{CDCl}_3$  at room temperature. In FIG. 2D, the  $^{13}\text{C}$  signals of the hydroxyl-bearing carbon of 2,2'-12-BTBA and 2,3-12-BTBA respectively undergo a significant downfield shift of approximately 7.0 and 7.5 ppm relative to the signals of both BTBAs without dense aggregation in 12-No-EDF. The  $\Delta\delta$  7.0 and 7.5 ppm downfield shifts are typical for the hydroxyl-bearing carbons of polyvinyl alcohol and are characteristic of the formation of

oligomeric alcohol clusters through continuous alcohol O—H—O—H hydrogen bonding (Ohgi, H., Sato, T., Hu, S. H. & Horii, F. Highly Isotactic Poly(Vinyl Alcohol) Derived From tert-Butyl Vinyl Ether. Part IV. Some Physical Properties, Structure And Hydrogen Bonding Of Highly Isotactic Poly(Vinyl Alcohol) Films. *Polymer* 47, 1324-1332, 2006).

To determine whether the acyl migration is caused by the sulfate radical or the basicity of the sulfate,  $(\text{NH}_4)_2\text{S}_2\text{O}_8$  was replaced by potassium carbonate ( $\text{K}_2\text{CO}_3$ ) to treat 12-No-EDF under the same reaction conditions, i.e.  $150^\circ\text{C}$ . for 38 hours. The  $^1\text{H}$  NMR spectra of 12-No-EDF, product obtained by treating 12-No-EDF with  $(\text{NH}_4)_2\text{S}_2\text{O}_8$ , and product obtained by treating 12-No-EDF with  $\text{K}_2\text{CO}_3$  were respectively shown in FIG. 3B, and the marks of hydrogen atoms of 2,2'-12-BTBA, 2,3-12-BTBA, free 2,2'-12-BTBA, free 3,3'-12-BTBA, and free 2,3-12-diacylglycerol-bisphenol-glycerol are shown in FIG. 3A. In spectrum 3a, it showed that 12-No-EDF is composed of 2,2'-12-BTBA (peak b2) and 3,3'-12-BTBA (peak b1). In spectrum 3b, it can be seen that the 12-EDFONP contains 2,2'-12-BTBA (peak b5) and 2,3'-12-BTBA (peak b3 and b4) with collective proton motion. In addition, a small amount of 2,3-diacylglycerol-bisphenol-glycerol (peak b6) was obtained, but no free 2,2'-isomer (peak b2) was observed. In spectrum 3c, it can be seen that no peaks b3 and b4 was obtained. Therefore, the radical-catalyzed 3,2-acyl migration was established.

12-EDFONP:  $^1\text{H}$  NMR (600 MHz,  $\text{CDCl}_3$ ):  $\delta$  7.27 (b, 4H), 7.11 (b, 4H), 6.88 (b, 4H), 5.73 (s, 0.04H), 5.52 (s, 1.2H), 5.42 (s, 0.02H), 4.78 (m, 0.04H), 4.66 (m, 0.04H), 4.52 (b, 1.4H), 4.36 (b, 0.3H), 4.11 (b, 18.96H), 1.76 (m, 12H), 1.60 (s, 6H), 1.46 (m, 12H), 1.27 (m, 96H), 0.87 (t, 18H,  $J=6.6$  Hz);  $^{13}\text{C}$  NMR (150 MHz,  $\text{CDCl}_3$ ):  $\delta$  170.9, 160.1, 143.8, 143.1, 127.8, 124.2, 114.4, 108.2, 77.1, 73.5, 71.8, 70.2, 69.2, 68.9, 68.7, 68.9, 67.9, 66.3, 64.4, 63.3, 62.3, 41.8, 31.9, 31.1, 30.4, 29.6, 26.1, 22.7, 14.1; Elementary analysis (calcd., found for  $\text{C}_{107}\text{H}_{180}\text{O}_{14}$ ): C (76.02, 76.74), H (10.73, 10.74).

14-EDFONP:  $^1\text{H}$  NMR (600 MHz,  $\text{CDCl}_3$ ):  $\delta$  7.27 (b, 4H), 7.11 (b, 4H), 6.88 (b, 4H), 5.73 (s, 0.04H), 5.52 (s, 1.2H), 5.42 (s, 0.02H), 4.78 (m, 0.04H), 4.66 (m, 0.04H), 4.52 (b, 1.4H), 4.36 (b, 0.3H), 4.11 (b, 18.96H), 1.76 (m, 12H), 1.60 (s, 6H), 1.46 (m, 12H), 1.27 (m, 120H), 0.87 (t, 18H,  $J=6.6$  Hz);  $^{13}\text{C}$  NMR (150 MHz,  $\text{CDCl}_3$ ):  $\delta$  170.9, 160.1, 143.8, 143.1, 127.8, 124.2, 114.4, 108.2, 77.1, 73.5, 71.8, 70.2, 69.2, 68.9, 68.7, 68.9, 67.9, 66.3, 64.4, 63.3, 62.3, 41.8, 31.9, 31.1, 30.4, 29.6, 26.1, 22.7, 14.1; Elementary analysis (calcd., found for  $\text{C}_{119}\text{H}_{204}\text{O}_{14}$ ): C (76.89, 77.34), H (11.06, 11.07).

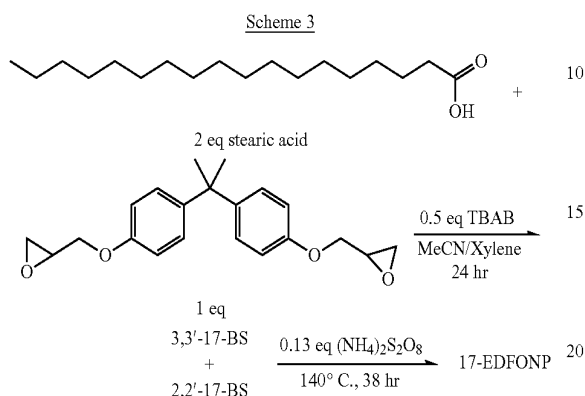
16-EDFONP:  $^1\text{H}$  NMR (600 MHz,  $\text{CDCl}_3$ ):  $\delta$  7.27 (b, 4H), 7.11 (b, 4H), 6.88 (b, 4H), 5.73 (s, 0.04H), 5.52 (s, 1.2H), 5.42 (s, 0.02H), 4.78 (m, 0.04H), 4.66 (m, 0.04H), 4.52 (b, 1.4H), 4.36 (b, 0.3H), 4.11 (b, 18.96H), 1.76 (m, 12H), 1.60 (s, 6H), 1.46 (m, 12H), 1.27 (m, 144H), 0.87 (t, 18H,  $J=6.6$  Hz);  $^{13}\text{C}$  NMR (150 MHz,  $\text{CDCl}_3$ ):  $\delta$  170.9, 160.1, 143.8, 143.1, 127.8, 124.2, 114.4, 108.2, 77.1, 73.5, 71.8, 70.2, 69.2, 68.9, 68.7, 68.9, 67.9, 66.3, 64.4, 63.3, 62.3, 41.8, 31.9, 31.1, 30.4, 29.6, 26.1, 22.7, 14.1; Elementary analysis (calcd., found for  $\text{C}_{131}\text{H}_{228}\text{O}_{14}$ ): C (77.61, 77.91), H (11.34, 11.38).

18-EDFONP:  $^1\text{H}$  NMR (600 MHz,  $\text{CDCl}_3$ ):  $\delta$  7.27 (b, 4H), 7.11 (b, 4H), 6.88 (b, 4H), 5.73 (s, 0.04H), 5.52 (s, 1.2H), 5.42 (s, 0.02H), 4.78 (m, 0.04H), 4.66 (m, 0.04H), 4.52 (b, 1.4H), 4.36 (b, 0.3H), 4.11 (b, 18.96H), 1.76 (m, 12H), 1.60 (s, 6H), 1.46 (m, 12H), 1.27 (m, 168H), 0.87 (t, 18H,  $J=6.6$  Hz);  $^{13}\text{C}$  NMR (150 MHz,  $\text{CDCl}_3$ ):  $\delta$  170.9, 160.1, 143.8, 143.1, 127.8, 124.2, 114.4, 108.2, 77.1, 73.5,

## 11

71.8, 70.2, 69.2, 68.9, 68.7, 68.9, 67.9, 66.3, 64.4, 63.3, 62.3, 41.8, 31.9, 31.1, 30.4, 29.6, 26.1, 22.7, 14.1; Elementary analysis (calcd., found for  $C_{143}H_{252}O_{14}$ ): C (78.23, 78.62), H (11.57, 11.57).

Embodiment 2: Synthesis of 17-No-EDF and 17-EDFONP 5



The synthesis of 17-No-EDF and 17-EDFONP are shown in Scheme 3. The detailed synthesis of 17-No-EDF and 17-EDFONP is described below.

17-No-EDF: A solution of bisphenol A diglycidyl ether (2.34 g, 6.87 mmol) and octadecanoic acid (3.91 g, 13.74 mmol) in mix-solvent of acetonitrile (40 ml) and xylene (20 ml) was reflux at 85° C. for 24 h. In this step, 0.5 equivalent of tetra-n-butylammonium bromide (TBAB) was added to increase the reactivity of epoxy ring opening reaction. The solvent was removed under reduced pressure. The crude product was recrystallized twice from acetonitrile, and a white solid of 17-No-EDF was obtained (4.81 g, 77%). The 17-No-EDF was composed of 2,2'-17-BS and 3,3'-17-BS positional isomers. The chemical structures of 2,2'-17-BS and 3,3'-17-BS are shown in FIG. 4A, and the  $^1H$  NMR spectrum of 17-No-EDF is shown in spectrum 4a of FIG. 4B. Some spectra data are described below.

$^1H$  NMR (400 MHz,  $CDCl_3$ ):  $\delta$  7.11 (d, 4H, J=8.4 Hz), 6.79 (d, 4H, J=8.4 Hz), 5.18 (d, 0.31H, J=6.8 Hz), 4.27 (m, 2.97H), 4.20 (m, 2H), 4.06 (d, 0.82H, J=5.2 Hz), 3.96 (m, 3.32H), 3.96 (d, 0.58H, J=5.2 Hz), 2.33 (t, 4H, J=7.6 Hz), 1.61 (b, 10H), 1.23 (b, 56H), 0.86 (t, 6H, J=6.8 Hz);  $^{13}C$  NMR (100 MHz,  $CDCl_3$ ):  $\delta$  174.0, 156.2, 143.8, 127.8, 114.0, 72.7, 68.7, 66.2, 65.2, 62.1, 41.7, 34.3, 33.6, 31.9, 31.0, 29.4, 24.9, 22.7, 14.1; MS (MALDI-TOF): 931.83  $[M+Na]^+$ .

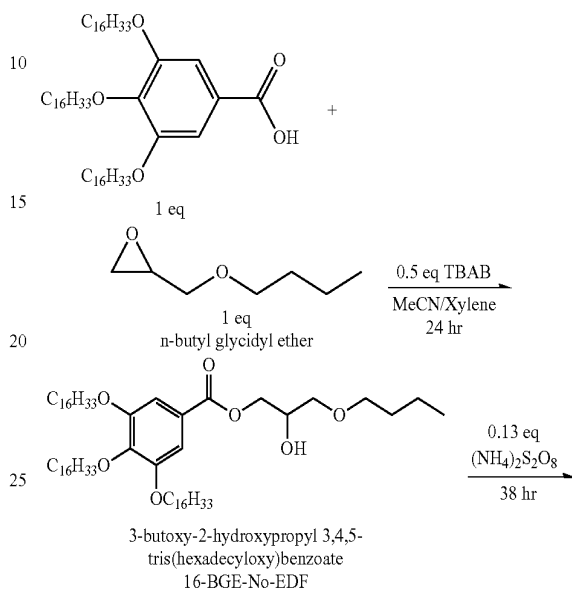
17-EDFONP: The 17-No-EDF (2.21 g, 0.32 mmol) and ammonium persulfate (0.07 g, 0.32 mmol) were heated and stirred under  $N_2$  atmosphere at 140° C. for 38 h. The crude product was then dissolved in  $CH_2Cl_2$  and filtered. The filtrate was dried under vacuum to give a brown product of 17-EDFONP with a yield of 64% (4.00 g). The 17-EDFONP was composed of 2,2'-17-BS, 2,3-17-BS, 3,3'-17-BS and 2,3-17-diacylglycerol-bisphenol-glycerol isomers, and no any non-acyl migrated side product is found. The chemical structures of 2,2'-17-BS, 2,3-17-BS, 3,3'-17-BS and 2,3-17-diacylglycerol-bisphenol-glycerol are shown in FIG. 4A, and the  $^1H$  NMR spectrum of 17-EDFONP is shown in spectrum 4b of FIG. 4B. Some spectra data are described below.

$^1H$  NMR (400 MHz,  $CDCl_3$ ):  $\delta$  7.09 (s, 4H), 6.77 (d, 4H, J=7.2 Hz), 5.35 (m, 1H), 5.25 (b, 0.8H), 4.42-3.60 (m, 8.2H), 2.34-2.27 (m, 4H), 1.50-1.60 (m, 10H), 1.15-1.30 (m, 56H), 0.86 (t, 6H, J=6.0 Hz);  $^{13}C$  NMR (100 MHz,  $CDCl_3$ ):  $\delta$

## 12

173.2, 156.1, 143.5, 127.7, 113.9, 84.0, 70.6, 69.5, 68.7, 66.2, 62.4, 47.4, 41.7, 34.3, 34.1, 31.1, 29.3, 24.9, 22.7, 14.1. Embodiment 3: Synthesis of 16-BGE-No-EDF and 16-BGE-EDFONP

Scheme 4



The synthesis of 16-BGE-No-EDF and 16-BGE-EDFONP are shown in Scheme 4. The detailed synthesis of 16-BGE-No-EDF and 16-BGE-EDFONP is described below.

16-BGE-No-EDF: A solution of butyl glycidyl ether (5.36 g, 41.20 mmol) and 3,4,5-tris(hexadecyloxy)benzoic acid (3.47 g, 4.12 mmol) in mix-solvent of acetonitrile (50 ml) and xylene (5 ml) was reflux for 24 h. The solvent was removed under reduced pressure and the crude product was purified by silica gel column chromatography using hexane and EtOAc as eluent to afford pure 3-butoxy-2-hydroxypropyl-3,4,5-tris(hexadecyloxy)benzoate (i.e. 16-BGE-No-EDF) in 55% combined yield (2.13 g). In this embodiment, since the steric hinderance is too large, only 3-monoacyl glycerol was obtained. The  $^1H$  NMR spectra of 16-BGE-No-EDF (spectrum 5a) is shown in FIG. 5B. Other spectrum data are described below.

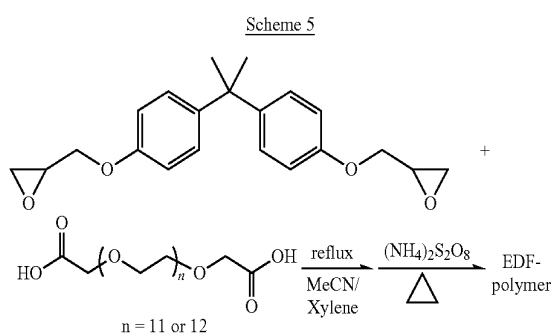
$^1H$  NMR (400 MHz,  $CDCl_3$ ):  $\delta$  7.24 (s, 2H), 4.35 (m, 2H), 4.09 (m, 1H), 3.98 (m, 6H), 3.50 (m, 4H), 1.81 (d, 1H, J=6.4 Hz), 1.75 (m, 6H), 1.55 (m, 2H), 1.45 (m, 6H), 0.88 (m, 12H);  $^{13}C$  NMR (100 MHz,  $CDCl_3$ ):  $\delta$  170.6, 166.6, 152.8, 143.0, 142.7, 124.3, 123.8, 108.5, 108.3, 73.8, 73.5, 71.5, 71.4, 70.0, 69.1, 66.2, 62.9, 31.9, 31.6, 30.3, 29.5, 26.1, 22.6, 19.2, 14.0, 13.8; MS (MALDI-TOF): 995.96  $[M+Na]^+$ .

16-BGE-EDFONP: The previous 16-BGE-No-EDF product (0.64 g, 0.65 mmol) and ammonium persulfate (0.02 g, 0.09 mmol) were heated and stirred under  $N_2$  at 140° C. for 38 h. The crude product was then dissolved in  $CH_2Cl_2$  and filtered. The filtrate was dried under vacuum to give a brown solid with a yield of 96% (0.61 g) which is composed of 3-butoxy-2-hydroxypropyl-3,4,5-tris(hexadecyloxy)benzoate (i.e. 3-monoacyl glycerol), 1-butoxy-3-hydroxypropan-2-yl 3,4,5-tris(hexadecyloxy)benzoate (i.e. 2-monoacyl glycerol), 3-butoxypropane-1,2-diyl bis(3,4,5-tris(hexadecyloxy)benzoate) (i.e. 2,3-diacyl glycerol) and 3-butoxypro-

pane-1,2-diol (i.e. no acyl substituent). The chemical structures of the components of the 16-BGE-EDFONP are shown in FIG. 5A. The  $^1\text{H}$  NMR spectra of 16-BGE-EDFONP (spectrum 5b) are shown in FIG. 5B. Some spectra data of the 16-BGE-No-EDF are described below.

$^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ ):  $\delta$  7.22 (m, 2H), 5.53 (b, 0.08H), 5.33 (m, 0.48H), 5.18 (b, 0.02H), 4.65 (m, 0.16H), 4.39 (m, 0.72H), 4.12 (m, 0.17H), 3.97 (m, 6.94H), 3.76 (m, 0.71H), 3.66 (m, 1.09H), 3.56 (m, 1.07H), 3.41 (m, 1.56H), 1.78 (m, 6H), 1.40 (m, 82H), 0.86 (t, 6H,  $J=7.2$  Hz);  $^{13}\text{C}$  NMR (100 MHz,  $\text{CDCl}_3$ ):  $\delta$  166.6, 152.8, 142.6, 124.3, 108.2, 76.6, 73.5, 71.6, 71.4, 70.1, 69.2, 69.0, 31.9, 31.6, 30.3, 29.5, 26.0, 22.6, 19.2, 14.1, 13.8.

Embodiment 4: Synthesis of EDF-Polymer



In Scheme 5, bisphenol A diglycidyl ether was used to react with poly(ethylene glycol)bis(carboxymethyl) ether, and an alternating copolymer (abbreviated as EDF-polymer) was obtained. The detailed synthesis of EDF-polymer is described below.

A solution of bisphenol A diglycidyl ether (3.02 g, 8.87 mmol) and poly(ethylene glycol)bis(carboxymethyl) ether with a number-average molecular weight ( $M_n$ ) about 600 (5.11 g, 8.85 mmol) in mix-solvent of acetonitrile (40 ml) and xylene (20 ml) was reflux at  $85^\circ\text{C}$ . for 24 h. The solvent was removed under reduced pressure. The crude product was recrystallized twice from acetonitrile, and a gel-type product is obtained (6.82 g, 82%).

Gel-type product (3.14 g) and ammonium persulfate (0.10 g, 0.43 mmol) were heated and stirred under  $\text{N}_2$  at  $150^\circ\text{C}$ . for 38 h. The crude product was then dissolved in  $\text{CH}_2\text{Cl}_2$  and filtered. The filtrate was dried under vacuum to give a brown product of a EDF-polymer with a yield of 92% (2.89 g).

The molecular weight of the EDF-Polymer was estimated by gel permeation chromatography and the number average molecular weight ( $M_n$ ) of the EDF-Polymer was approximately 12,000 Da. The EDF-polymer contained 2-monoacylglycerol, 3-monoacylglycerol and diacylglycerol moieties, and no any non-acyl migrated side product was found. The chemical structures of the EDF-polymers is shown in FIG. 6A. The  $^1\text{H}$  NMR spectrum of the EDF-polymer in  $\text{CDCl}_3$  at room temperature is shown in FIG. 6B. Some spectra data are described below.

$^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ ):  $\delta$  7.09 (d,  $J=8.0$  Hz), 6.77 (d,  $J=8.0$  Hz), 5.41 (b), 5.27 (m), 4.71 (b), 4.51 (b), 4.36-4.04 (m), 3.97 (b), 3.84 (b), 3.78 (b), 3.61 (b), 3.44 (b), 3.31 (b), 2.81 (m), 2.72 (b), 1.78-1.74 (m, 12H), 1.60 (b);  $^{13}\text{C}$  NMR (100 MHz,  $\text{CDCl}_3$ ):  $\delta$  171.5, 156.3, 143.5, 113.9, 77.3, 73.5, 72.5, 71.8, 71.7, 70.9, 69.1, 68.0, 65.9, 63.7, 63.0, 61.6, 58.8, 41.7, 31.0.

## Characterization and Properties of EDFONPs

Investigations based on the combination of NMR, high-resolution transmission electron microscopy (HRTEM), dynamic light scattering (DLS) and photoluminescence (PL) spectra reveal that the densely aggregated structures of the acyl-migrated product are discrete self-assembled excitation-dependent fluorescent organic nanoparticles (EDFONPs).

### Particle Size Measurements

The HRTEM image of 18-EDFONP with a diameter of approximately 7.6 nm is shown in FIG. 7A. The HRTEM images were taken with a JEM-2100F (JEOL, Japan) operated at 200 kV, and the sample was prepared by drop-casting a suspension of  $\text{CH}_2\text{Cl}_2$  onto a quantifoil holey carbon grid.

The mean diameters, measured by dynamic light scattering, of 12-EDFONP to 18-EDFONP in  $\text{CH}_2\text{Cl}_2$  are shown in FIG. 7B. The hydrodynamic particle size is measured with dynamic light scattering (Brookhaven 90 Plus, USA). The obtained mean diameters of 12-EDFONP to 18-EDFONP are 4.8, 6.0, 8.6 and 9.5 nm, respectively.

FIG. 7C is a HRTEM image of the EDF-polymer above shows aggregated nanoparticles with a scale bar of 10 nm. It shows that the diameter of the nanoparticles of the EDF-polymer is about 5 nm.

### Excitation and Emission Spectrum

All of the EDFONPs above have similar photoluminescence excitation and emission behaviors in  $\text{CH}_2\text{Cl}_2$  and  $\text{CHCl}_3$  solvents. FIG. 8A is excitation and emission spectra of 12-EDFONP at a concentration of 10 mM. In FIG. 8A, the excitation spectrum of 12-EDFONP exhibits a broad peak at 330 nm with a shoulder that extends to approximately 650 nm. The excitation of 12-EDFONP from 360 to 600 nm results in an excitation-dependent fluorescence from 415 to 660 nm, where the intensity remarkably decreases. FIG. 8B is a fluorescent photograph of 12-EDFONP in a  $\text{CH}_2\text{Cl}_2$  solution, which was simultaneously excited at 404 and 510 nm and exhibits intense blue and yellow fluorescence in the same quartz cuvette.

FIG. 9A is excitation and emission spectra of 12-No-EDF at concentrations of 0.01 and 10 mM. It can be clearly seen that no excitation dependent fluorescence occurred. In FIG. 9A, the excitation spectrum of 12-No-EDF at a concentration of 0.01 mM and 10 mM displays only one excitation peak at 274 nm and 329 nm, respectively. The emission spectra of 12-No-EDF at a concentration of 0.01 mM and 10 mM were the same, and had a relatively shaper UV fluorescence peak at 353 nm when both were excited at 274 nm and 329 nm, respectively.

FIGS. 9B and 9C are excitation and emission spectra of 12-EDFONP at an initial concentration of 10 mM (red line), then diluted to 0.01 mM (blue line) and reconcentrated back to 10 mM (green line), respectively. Comparing FIGS. 9A to 9C, when the concentrations of 12-EDFONP and 12-No-EDF are subsequently decreased from 10 to 0.01 mM by dilution, both exhibit similar excitation and emission spectra. Two excitation peaks at 235 and 274 nm and one emission peak at 355 nm (excited at 274 nm) were observed, which indicated that both 12-EDFONP and 12-No-EDF dissociated into individual 3,3'-12-BTBA and 2,2'-12-BTBA molecules without dense aggregation when diluted to a concentration of 0.01 mM.

However, when 12-EDFONP is reconcentrated from 0.01 mM to 10 mM, the broad characteristic excitation (spectrum in green in FIG. 9B) and excitation-dependent fluorescence reappears (spectrum in FIG. 9C). This result clearly indicates that the EDF behavior is induced by the dense aggre-

15

gation in the self-assembled organic nanoparticles but not by the individual 12-BTBA molecule itself.

To determine the functional groups—including bisphenol, benzoate and monoacylglycerol—that are responsible for the EDF behavior, two compounds were prepared by changing the benzoate and bisphenol A groups to the heptadecyl carboxylate and n-butyl glycidyl ether groups, respectively. The obtained EDFONPs are 17-EDFONP and 16-BGE-EDFONP above.

FIG. 10 is emission spectrum of 17-EDFONP based on stearic acid at a concentration of 10 mM with a visible EDF phenomena. FIG. 11 is emission spectrum of 16-BGE-EDFONP at a concentration of 10 mM with a visible EDF phenomena. From FIGS. 10 and 11, it can be known that both of the modified compounds also exhibited the EDF behavior, which clearly indicates that the 2-monoacylglycerol group is responsible for the EDF behavior. In addition, this result also indicates that the EDF behavior is a common phenomenon for the 2-monoacylglycerol-dominated mixture. Note that the bis(monoacyl glycerol) clusters serve as chromophores here without any extended conjugated system.

FIG. 12 is emission spectrum of the EDF-Polymer. In FIG. 12, the emission spectrum of EDF-Polymer in  $\text{CHCl}_3$  also exhibited an excitation-dependent fluorescence behavior which is similar to that of the n-EDFONPs.

#### Quantum Yield and Fluorescence Lifetime

The quantum yield of 12-EDFONP and the EDF-polymer was respectively determined to be 8.3% and 6.1% with coumarin 1 (i.e. 7-diethylamino-4-methylcoumarin) in ethanol (quantum yields=73%) as a reference. Quantum Yields are calculated using the equation (1) below, where I is integrated fluorescence intensity, E is extinction coefficient, n is refractive index, and the subscript index R indicates the standard.

$$Q = (Q_R I E_R n^2) / (I_R E n_R^2) \quad (1)$$

The fluorescence lifetime measurement of the excitations of 12-No-EDF at 329 nm and 12-EDFONP at 360 nm are determined to be 0.98 and 1.54 ns, respectively.

#### Structural Characterization of the Self-Assembled EDFONPs

Further structural characterization using NMR experiments indicate the formation of decamers for n-BTBA. The decamer contains five 2,2'-n-BTBA and five 2,3-n-BTBA Diffusion Coefficient Measurement

The diffusion coefficient is measured using the NMR DOSY technique for solutions of 18-EDFONP, and the result is shown FIG. 13. In FIG. 13, the spectrum shows only one diffusion species that is characterized by the diffusion coefficient (D) of  $0.95 \times 10^{-10} \text{ m}^2 \text{ s}^{-1}$ .

The hydrodynamic radius ( $r_H$ ) of 3.9 nm is calculated from the Stokes-Einstein equation (2) below, where k is the Boltzmann constant, T is the temperature, and  $\eta$  is the viscosity of solution.

$$r_H = kbT / 6\pi\eta D \quad (2)$$

The molecular weight of 18-EDFONP is estimated to be approximately 24,000 from the Mark-Houwink-Sakaruda (MHS) equation (3) below, which is determined by the phenolphthalein poly(ether sulfone) in chloroform solution.

$$D = (2.45 \pm 0.04) \times 10^{-4} \text{ MW}^{-(0.55 \pm 0.05)} \quad (3)$$

16

Therefore, it was suggested that the nanoparticles of 18-EDFONP contain clusters having about ten 18-BTBA.

#### Components and Geometry of the Decamer

The components of the decamer was determined by  $^1\text{H}$  NMR. From the  $^1\text{H}$  NMR of 12-EDFONP (see FIG. 2A), the ratio of 2,2'-12-BTBA and 2,3-12-BTBA determined using  $^1\text{H}$  NMR is approximately 1:1. Therefore, the decamer should be composed of five 2,2'-n-BTBA and five 2,3-n-BTBA, i.e. there were fifteen 2-monoacylglycerol and five 2-monoacylglycerol. These 20 monoacylglycerol moieties could construct a Great Stellated Dodecahedral, in which every three neighboring monoacylglycerol groups form a triangular unit. The formation of Great Stellated Dodecahedral decamers by the self-assembly of five 2,2'-n-BTBA and five 2,3-n-BTBA is shown in FIG. 14. In FIG. 14, the 2-positional isomers are represented by red rods, and the 3-positional isomers are represented by blue rods.

#### Geometry of EDF-polymer

FIG. 15 is a cartoon model of nanoparticle of EDF-polymer. In FIG. 15A, a nanoparticle of EDF-Polymer are constructed through a self-assemble process with that the bisphenol A aggregates to serve as a core (blue parts), and the poly(ethylene glycol) moieties serves as a shell (red parts).

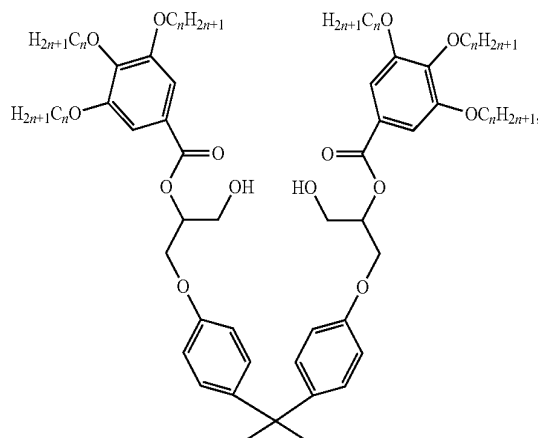
According to the results mentioned above, the EDFONPs can be formed through a ring opening reaction and a radical-catalyzed 3,2-acyl migration between an epoxy glycidyl ether and a carboxylic acid.

All the features disclosed in this specification (including any accompanying claims, abstract, and drawings) may be replaced by alternative features serving the same, equivalent or similar purpose, unless expressly stated otherwise. Thus, each feature disclosed is one example only of a generic series of equivalent or similar features.

What is claimed is:

1. Excitation-dependent fluorescent organic nanoparticles, each comprising:

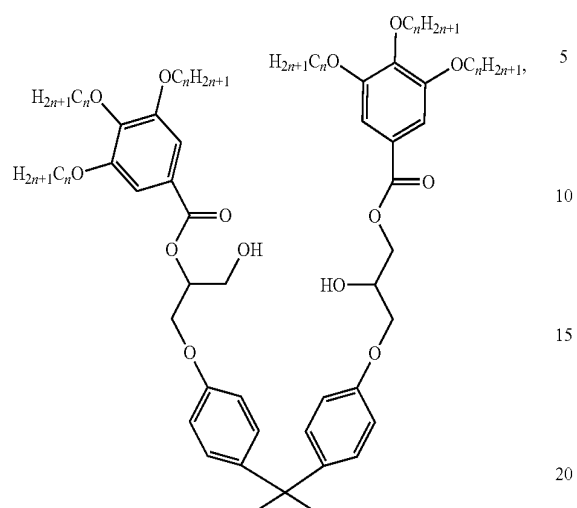
a cluster of aggregated molecules of 2-monoacylglycerol and 3-monoacylglycerol, wherein 2-monoacylglycerol comprises



17

3-monoacylglycerol comprises

18

and  $n=12-18$ .

\* \* \* \* \*