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(54) Title: DIODE-LASER COMPATIBLE AZA-DIPYRROMETHENE DYES AND METHODS FOR THEIR PREPARATION

(57) Abstract: The present invention relates to azadipyrromethene compounds, particularly azadipyrromethene metal complexes, pharmaceutical compositions comprising said compounds, as well as methods for their preparation and their uses, such as in photodynamic therapy.

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Diode-Laser Compatible Aza-dipyrromethene Dyes and Methods for Their Preparation

Field of the Invention

The present invention relates to azadipyrromethene compounds, particularly azadipyrromethene metal complexes, pharmaceutical compositions comprising said compounds, as well as methods for their preparation and their uses, such as in photodynamic therapy.

Background of the Invention

Near-infrared (NIR) dyes are important in various applications such as optical recording, thermal writing display, laser printing, bio sensing, photodynamic therapy (PDT) and the like (Fabian, M.; Nakazumi, H.; Matsuka M. *Chem. Rev.* **1992**, 1197-1226).

The principal advantage of working in the NIR region (650-1100 nm) is the absence or significant reduction of background signals (absorption, fluorescence, and light scatting) that are prevalent in the UV/Visible region of the spectrum. Another advantage is the wide availability of low-cost diode lasers as source of irradiation. As bio sensing by fluorescence is concerned, very few biological molecules posses intrinsic fluorescence in the NIR region. Light scatting is relevant to $1/\lambda^4$, which results in much less reduction of scatting in NIR region. As a result, the detection limit for the NIR dyes is a couple of orders lower than visible dyes (Sowell, J.; Strekowski, L.; Patonay, G. J. *Biomed. Opt.* **2002**, *7*, 571-575). As PDT is concerned, whereby a photosensitising compound, which is administered to a target tissue and light-activated by illumination of the tissue with light of a wavelength

absorbed by the compound, transfers its excited state energy to the surrounding biological tissue, resulting in oxidative cellular damage and cell death, the light penetration through tissues up to 10-20 cm is feasible in the NIR region (Ntziachristos, V.; Ripoll, J.; Weissleder, R. *Opt. Lett.* **2002**, *27*, 333-335).

A number of dyes that have previously been found to be fluorescent do not have significant absorbance in NIR region. The most common NIR fluorescent dyes used for bio sensing are polymethines, especially heptamethine cyanines (Frnagioni, J. V. *Curr. Opi. Chem. Bio.* **2003**, *7*, 626-634). However, the NIR cyanine dyes are known to be fatigue-prone and weakly fluorescent (Mishra, A.; Behera, R. K.; Behera, P. K.; Mishra, B. B.; Behera, G. B. *Chem. Rev.* **2000**, *100*, 1973-2011), and may form aggregate which quenches fluorescence.

The haematoporphyrin derivative Photofrin® is the most commonly used clinically available PDT agent which has been approved for use in the United States, Japan and Europe. However Photofrin® and other porphyrin PDT agents have low absorption in the therapeutic window (650-800 nm). The second generation of PDT agents under investigation is phthalo- or naphthalocyanines. However they suffer from the drawback that they have very low solubility in most cases.

Recently, specific azadipyrromethene dyes, which were first reported in 1940's (US 2,469,830), have also been disclosed for various uses, including their use in optical recording (JP 11092479 and JP 11034500), in bio sensing (US 5,786,219, US 5,723,218, US 5,573,909 and WO 93/23492) and as PDT agents (WO 03/080627). However, these reported dyes generally absorb only below 700 nm with relatively high extinction coefficient (~80000 $M^{-1}cm^{-1}$).

Also, many general problems have been encountered with the synthesis of NIR fluorochromes compared to visible light fluorochromes. These include (1) significant spectral broadening as the wavelength increases, (2) low quantum yield, (3) photoinstability, (4) chemical instability with increasing red-shift, (5) the tendency to aggregate because of increased hydrophobicity, and (6) intramolecular electronic transfer may occur with more electron rich donor.

Thus there is a need for developing novel, easily accessible, fluorescent NIR dyes which overcome the above described disadvantages, i.e. NIR dyes characterized by having for example good stability, relatively high fluorescence quantum efficiency and ease of preparation.

Applicants have now surprisingly found that the novel diode-laser compatible azadipyrromethene dyes of the present invention are able to overcome some of the above described problems.

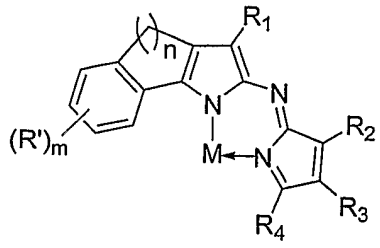
Brief Description of the Drawings

Fig. 1: The absorption spectra of compound **1** and reference compound **15** reported in WO 03/ 080627 in chloroform (1.0×10^{-5} mol/l). The absorption maximum of compound **1** occurs at 740 nm (extinction coefficient 159000 $M^{-1}cm^{-1}$) and half band width 30.4 nm; the reference dye absorbed at 688 nm (extinction coefficient 78500 $M^{-1}cm^{-1}$) and half band width 57 nm.

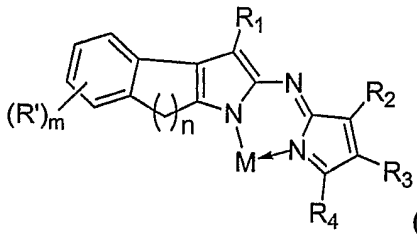
Fig. 2: The emission spectra of compound **1** and reference compound **15** reported in WO 03/080627 in chloroform excited at 650 nm. The Emission maximum of compound **1** occurs at 751 nm with quantum efficiency 0.28 relative to reference compound. The reference compound emitted at 715 nm with quantum efficiency of 0.36 in chloroform.

Detailed Description of the Invention and Preferred Embodiments

The present invention provides novel azadipyrromethene derivatives that exhibit narrow and sharp long wavelength absorption where the use of NIR is particularly advantageous. In one embodiment, the present invention provides a compound of general formula (I) or (II)



(I)



(II)

or a salt, metal complex or hydrate thereof

wherein

M is a chelating agent select from BF, BF_2, BCl, BCl_2, Zn, Mg, Al, Si, Sn, Cu, Ni, Co;

R' is hydrogen, hydroxy, halogen, nitro, cyano, allyl, linear or branched (C_1-C_{20})alkyl, (C_3-C_{20})cycloalkyl, (C_1-C_{20})alkoxy, (C_1-C_{20})alkylacetylenyl, phenylacetylenyl, (C_2-C_{20})alkenyl, phenylvinyl, halo(C_1-C_{20})alkyl, halo(C_3-C_{20})cycloalkyl, halo(C_1-C_{20})alkoxy, aryl, aryloxy or heteroaryl optionally substituted with (C_1-C_6)alkyl or (C_1-C_6)alkoxy; arylalkyl or heteroarylalkyl; nitrogen-containing heterocycloalkyl having 5 or 6 atoms optionally substituted with (C_1-C_6)alkyl or (C_1-C_6)alkoxy, $-N(R_A)R_B$, $CON(R_A)R_B$, wherein R_A and R_B are the same or different and are each independently hydrogen, (C_1-C_{20})alkyl, (C_3-C_{20})cycloalkyl, and optionally substituted phenyl; –OCOR, –COOR or –COR, wherein R represents hydrogen, (C_1-C_{20})alkyl, (C_3-C_{20})cycloalkyl, or aryl or heteroaryl optionally substituted with (C_1-C_6 carboxylate)alkyl or (C_1-C_6)alkoxy; or R' may also be a water solubilizing group such as sulfonate, carboxylate, or quaternary ammonium salt;

R_1-R_4 are the same or different and are each independently:

(a) linear or branched (C_1-C_{12})alkyl, (C_3-C_{12})cycloalkyl, aryl(C_1-C_6)alkyl or heteroaryl(C_1-C_6)alkyl, (C_1-C_6)alkoxy(C_1-C_6)alkyl, (C_1-C_{12})alkoxy, halo(C_1-C_{12})alkyl, (C_1-C_{12})haloalkoxy, (C_1-C_{12})alkylthio;

(b) substituted or unsubstituted aryl groups;

(c) substituted or unsubstituted heteroaryl groups;

(d) a group of the following formulae:

wherein B is hydrogen, (C_1-C_{12})alkyl or substituted or unsubstituted aryl;

(e) unsubstituted or mono-substituted pyrazolyl, pyridyl, imidazolyl, pyrazolinyl, imidazolinyl, or acridinyl, each of the said substituents being (C_1-C_6)alkyl, (C_1-C_6)alkoxy, fluoro, chloro, or phenyl.

(f) a group of the following formulae:

wherein C and D may be the same or different and are each independently carbon, oxygen, (C_1-C_{12})alkyl nitrogen, or (C_1-C_{12})acyl nitrogen;

R_C and R_D are each hydrogen or (C_1-C_{12})alkyl; and

wherein the phenyl moiety is optionally substituted with (C_1-C_{12})alkyl, (C_1-C_{12})alkoxy, (C_2-C_{12})acyl, fluoro, or chloro ;

(g) a water solubilizing group such as sulfonate, carboxylate, or quaternary ammonium salt;

(h) R_2 and R_3, R_3 and R_4 may be joined to form a ring with ring size of 5, 6 or 7;

n is an integer from 1 to 4; and

m is an integer from 0 to 4.

"Alkyl" whether used alone or when used as a part of another group includes straight and branched chain alkyl groups containing up to 20 atoms preferably up to

12 atoms, more preferably up to 8 atoms and most preferably up to 6 atoms such as methyl, ethyl, propyl, isopropyl, t-butyl, n-butyl, heptyl and hexyl.

"Cycloalkyl" includes a non-aromatic cyclic or multicyclic ring system of 3 to 20 carbon atoms, preferably 3 to 12, more preferably 3 to 8, and most preferably 5 or 6 carbon atoms. The cyclic alkyl may optionally be partially unsaturated.

"Alkoxy" means an alkyl-O- group in which the alkyl group is as defined above.

"Halogen" or "halo" means fluoro, chloro, bromo, or iodo, preferably fluoro, chloro, or bromo.

"Aryl" includes, without limitation, aromatic radicals containing from 6 to 10 carbon atoms, preferably phenyl or naphthyl.

"Aryloxy" means an aryl-O- group in which the aryl group is as defined above.

"Heteroaryl" includes a 5 to 10 membered aromatic monocyclic or multicyclic hydrocarbon ring system in which one or more of the atoms in the ring system is an element other than carbon, chosen from amongst nitrogen, oxygen or sulphur; if desired, a N atom may be in the form of an N-oxide. Preferred ring systems include without limitation, furyl, thienyl, pyrryl, indolyl, benzofuryl, benzothienyl, pyridyl, dibenzofuryl, dibenzothienyl, and carbazolyl, 2,2'-bithienyl, preferably furyl, thienyl, indolyl.

"Nitrogen-containing heterocyclic ring having 5 or 6 atoms" includes, without limitation, pyrrolidino, piperidino, morpholino, and the like.

Arylalkyl means an aryl-alkyl- group wherein the aryl and alkyl are as described above. Heteroarylalkyl means a heteroaryl-alkyl group, the heteroaryl and alkyl are as defined above.

"Substituted aryl or heteroaryl groups" includes, without limitation aryl or heteroaryl groups as defined above that are mono-, di-, or tri-substituted by a substituent that is: halogen nitro, amino, cyano, hydroxy, epoxy, vinyl, allyl, hydroxyethoxy, methoxyethoxy, hydroxyethoxyethoxy, methoxyethoxyethoxy;

$(C_1\text{-}C_{12})$alkyl, $(C_1\text{-}C_{12})$alkoxy, $(C_1\text{-}C_{12})$alkylaryl, aryl, aryloxy, aryl$(C_1\text{-}C_{12})$alkyl, aryl$(C_1\text{-}C_{12})$alkoxy, $(C_1\text{-}C_{12})$alkoxyaryl, halo$(C_1\text{-}C_{12})$alkyl, haloaryl, cyclo$(C_3\text{-}C_{12})$alkyl, cyclo$(C_1\text{-}C_{12})$alkoxy, aryloxyaryl, aryloxy$(C_1\text{-}C_{12})$alkyl, aryloxy$(C_1\text{-}C_{12})$alkoxy, acryloxy, methacryloxy; a heterocyclic nitrogen-containing substituent, such as N-$(C_1\text{-}C_{12})$alkylpiperazino, N-aryl-piperizino, aziridino, indolino, pyrrolidino, pyrrolino, piperidino, $(C_1\text{-}C_4)$alkylpiperidino, di$(C_1\text{-}C_4)$alkylpiperidino, 4-piperidinopiperidino, morpholino, 2,6-di$(C_1\text{-}C_4)$alkylmorpholino, thiomorpholino, thioazolidino, tetrahydroquinolino, pyrryl; $-N(R_A)R_B$, $CON(R_A)R_B$, wherein R_A and R_B are the same or different and are independently hydrogen, $(C_1\text{-}C_{12})$alkyl, $(C_3\text{-}C_{12})$cycloalkyl, phenyl, mono- or di-substituted phenyl; or –COR, –OCOR or –COOR, wherein R is hydrogen, $(C_1\text{-}C_{12})$alkyl, $(C_3\text{-}C_{12})$cycloalkyl, halo$(C_1\text{-}C_6)$alkyl, unsubstituted, mono- or di-substituted phenyl, unsubstituted, mono- or di-substituted naphthyl, unsubstituted, mono- or di- substituted furyl, or thienyl, and combination thereof.

In a preferred embodiment, the present invention provides compounds of formula (I) or (II) wherein:

M is BF, BF_2, Zn, Mg, Cu, Ni, Co;

R' is selected from hydrogen, nitro, cyano, allyl, fluoro, chloro, bromo, trifluoromethyl, trichloromethyl, dimethylamino, diethylamino, pyrrolidino, piperidino, morpholino, phenyl, benzyl; linear or branched $(C_1\text{-}C_6)$alkyl, $(C_1\text{-}C_6)$alkoxy, or –OCOR or –COOR wherein R is hydrogen, $(C_1\text{-}C_6)$alkyl, $(C_3\text{-}C_6)$cycloalkyl, or R' may also be a water solubilizing group such as sulfonate, carboxylate, or quaternary ammonium salt;

$R_1\text{-}R_4$ are the same or different and are each independently:

(a) linear or branched $(C_1\text{-}C_6)$alkyl, $(C_3\text{-}C_6)$cycloalkyl, aryl$(C_1\text{-}C_4)$alkyl or heteroaryl$(C_1\text{-}C_4)$alkyl, $(C_1\text{-}C_6)$alkoxy$(C_1\text{-}C_6)$alkyl;

(b) unsubstituted, mono-, di-substituted aryl selected from phenyl or naphthyl, preferably substituted in the ortho, the para position, or both;

(c) unsubstituted or mono- substituted heteroaryl groups that are furyl, thienyl, 2,2'-bithienyl, pyrryl, indolyl, benzofuryl, benzothienyl, pyridyl, dibenzofuryl, dibenzothienyl, or carbazolyl.

the substituents being amino, cyano, hydroxy, hydroxyethoxy, methoxyethoxy, hydroxyethoxyethoxy, methoxyethoxyethoxy, fluoro, chloro, bromo, iodo, vinyl, allyl, trifluoromethyl, phenyl, (C_1-C_6)alkyl, (C_1-C_6)alkoxy, cyclo(C_3-C_6)alkyl, cyclo(C_1-C_6)alkoxy, (C_1-C_6)alkylamino, di(C_1-C_6)alkylamino, diarylamino, phenylacetylenyl, or phenylvinyl;

a heterocyclic nitrogen-containing substituent, such as N(C_1-C_6)alkylpiperazino, N-aryl-piperizino, aziridino, indolino, pyrrolidino, pyrrolino, piperidino, (C_1-C_4)alkylpiperidino, di(C_1-C_4)alkylpiperidino, 4-piperidinopiperidino, morpholino, 2,6-di(C_1-C_4)alkylmorpholino, thiomorpholino, thioazolidino, tetrahydroquinolino, or pyrryl;

$N(R_A)R_B$, $CON(R_A)R_B$, wherein R_A and R_B are the same or different and are each independently hydrogen, (C_1-C_6)alkyl, (C_3-C_6)cycloalkyl, phenyl or –COR, –OCOR or –COOR wherein R is hydrogen, (C_1-C_6)alkyl, (C_3-C_6)cycloalkyl, or phenyl;

(d) a water solubilizing group such as sulfonate, carboxylate, or quaternary ammonium salt;

(e) R_2 and R_3, R_3 and R_4 may be joined to form a ring with ring size of 5, 6 or 7;

n is an integer from 1 to 3; and

m is an integer from 0 to 2.

In a still more preferred embodiment, the invention provides a compound of formula (I) or (II) wherein:

M is BF or BF_2;

R' is hydrogen, fluoro, chloro, bromo, dimethylamino, diethylamino, pyrrolidino, piperidino, morpholino, phenyl, benzyl, (C_1-C_4)alkyl, or (C_1-C_4)alkoxy, or R' may also be a water solubilizing group such as sulfonate, carboxylate, or quaternary ammonium salt;

R_1-R_4 are the same or different and are each independently:

unsubstituted, mono-, or di-substituted phenyl, preferably substituted in the alpha position, para position or both with the substituents being one or more of amino, acyl, cyano, methoxy, ethoxy, methoxyethoxy, fluoro, chloro, vinyl, allyl, methoxycarbonyl, ethoxycarbonyl, (C_1-C_4)alkyl, di(C_1-C_4)alkylamino, piperazino, piperidino, arylperidino, morpholino, pyrrolidino, aziridino, acryloxy, methacryloxy, phenylacetylenyl, phenylvinyl;

unsubstituted, mono-substituted heteroaromatic groups, such as furyl, thienyl, 2,2'-bithienyl, pyrryl, substituted with a substituent that is (C_1-C_4)alkyl or phenyl;

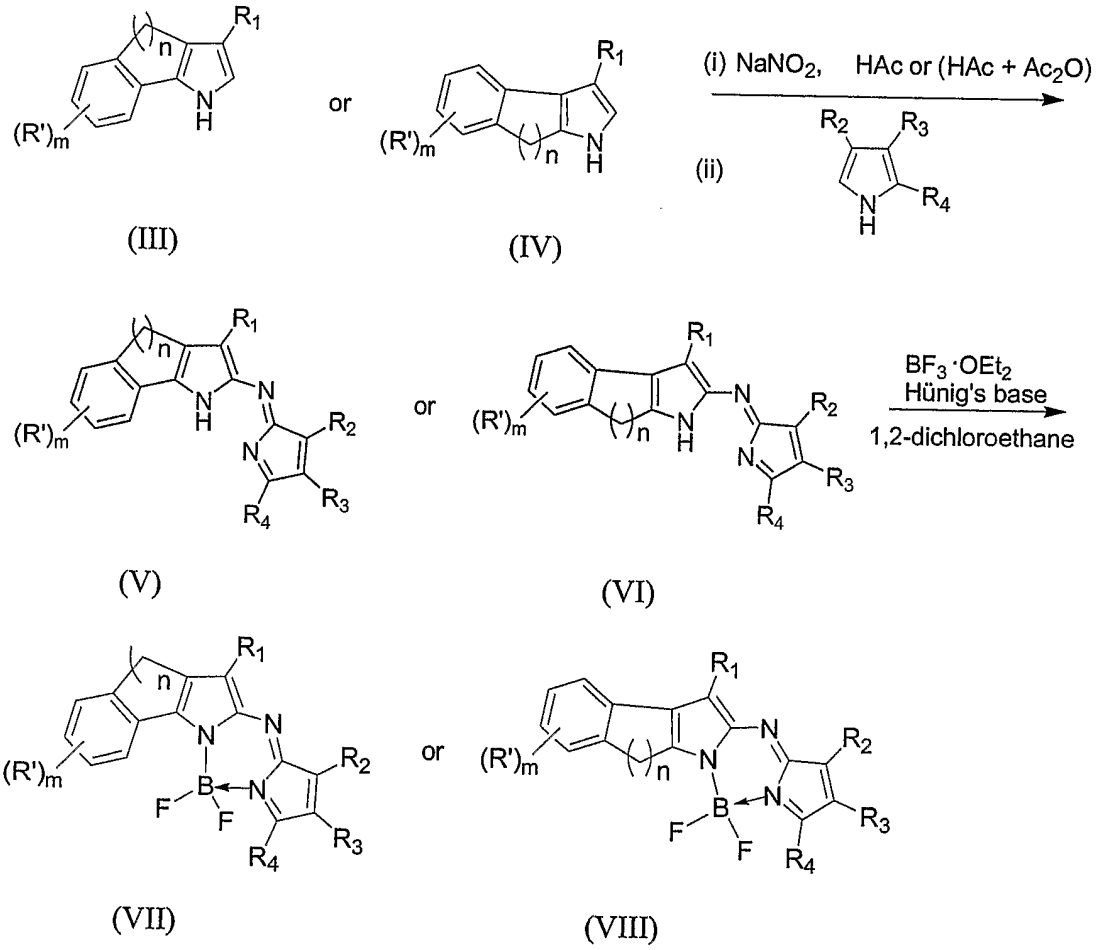
a water solubilizing group such as sulfonate, carboxylate, or quaternary ammonium salt;

R_2 and R_3, R_3 and R_4 may be joined to form a ring with ring size of 5, or 6;

n is an integer from 1 to 3, and

m is, independently, integer from 0 to 2.

A compound of formula (I) or (II), wherein M is BF_2 (formula (VII) or (VIII), respectively) may be prepared by a process shown below (Scheme 1):



(III) (IV)

(V) (VI)

(VII) (VIII)

Scheme 1

The most convenient synthesis of dipyrromethene was developed by Knott (*US*, **1949**, 2469830) by reaction of pyrrole (III) or (IV) derivatives with nitrosopyrrole which was prepared separately. In the present invention, applicants discovered that the nitrosopyrrole may be prepared *in situ* which allows the preparation of dipyrromethene intermediates (V) and (VI) (see Scheme 1) in one pot fashion. The reaction solvent may be selected from weak acid such as acetic acid, formic acid, propionic acid, chloroacetic acid, dichloroacetic acid, trichloroacetic acid, oxalic aicd. The reaction solvent may also be a mixture of the above mentioned

acid or a mixture of the acid with its anhydride or mixture of anhydrides. Preferably the reaction solvent may be acetic acid or acetic acid and acetic anhydride mixture. The nitrosating reagent in step (i) may be a solid or aqueous solution of metal nitrite. Said metal nitrite may be selected from sodium nitrite, lithium nitrite, potassium nitrite, ammonium nitrite, magnesium nitrite, calcium nitrite, or mixture of them. Preferably, said nitrite salt may be sodium nitrite, potassium nitrite, magnesium nitrite, more preferably sodium nitrite. The reaction temperatures may be from about -5 to about 160 °C, preferably about 0 to about 130 °C, more preferably about 0 to 80 °C. Reaction time may be about 1 minute to about 1 day, preferably about 10 minutes to about 2 hours, and more preferably about 20 minutes to about 1 hour.

Complex formation according to step (iii) may be carried out with trifluoroborane etherate in the presence of a suitable base in a non-polar solvent such as dichloromethane, chloroform, 1,2-dichloromethane, 1,1-dichloromethane, benzene, toluene, xylene; preferably 1,2-dichloromethane, benzene or toluene; more preferably 1,2-dichlormethane. Suitable base include, but are not limited to, triethylamine, Hünig's amine, diisopropylamine, piperidine and other similar bases. The reaction temperature may be from about 0 to about 130 °C, preferably about 20 to about 100 °C, more preferably about 20 to 80 °C.

One particular advantage of the compounds of the present invention is that fusion of the aromatic ring with pyrrole provides better conjugation which results in NIR absorption of the azadipyrromethene dye with high extinction coefficient and narrow half width, at the same time maintaining good fluorescence quantum yield.

The resulting products are generally soluble in organic solvents. Aqueous solubility can be obtained by adding appropriate water solubilization groups that include sulfonates, carboxylates, quaternary amine salts. In most cases, the dyes are easily purified by chromatography and/or recrystalization.

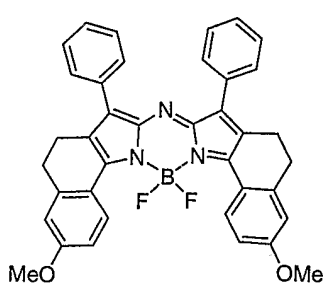
The highly fluorescent, diode laser compatible azadipyrromethene dyes of the invention may be found wide applications for fluorescent labeling, optical recording, and PDT. The reactive dye may also be conjugated to a bioactive molecule, which could be particular useful for the fluorescent labeling of members of specific binding pairs to form dye conjugates which can be used to detect biological interactions. The dye-conjugated ligands are useful in a wide variety of areas as biochemical tools for the detection, identification, and measurement of biological compounds. The dyes of the present invention and its conjugates may be particularly suitable for PDT due to the strong and sharp absorption in the therapeutic window where the body tissues have very low absorption. The dyes of the present invention may be modified by heavy atom which is well known to increase the population of excited triplet state to furnish better singlet oxygen generation. The azadipyrromethene of the present invention may form complex with other metal such as Ni, Cu, or Co, which is widely known to be non-fluorescent may find applications in optical recording.

Table 1 shows the chemical structure and absorption maxima in dichloromethane of the selected embodiments of the dyes of the present invention and a reference dye (**15**) in the patent application WO 03/080627.

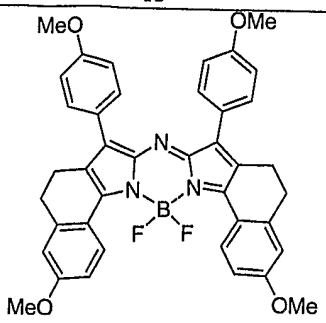
The dyes of the invention have a sharp absorption and possess an absorption maximum at a wavelength of 660 nm or longer, preferably at a wavelength of 720 nm or longer. Further, the dyes of the invention posses an emission maximum at a wavelength of 700 nm or longer, preferably, 740 nm or longer.

Table 1

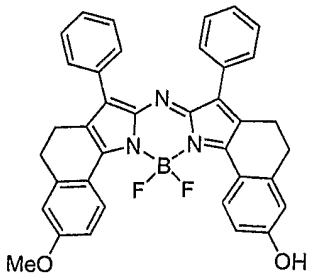
Selected Dyes of the Present Invention



1 (λ_{max}: 740 nm)



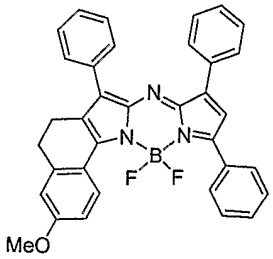
2 (λ_{max}: 737 nm)



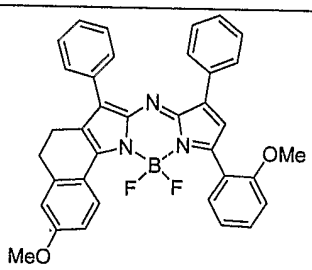
3 (λ_{max}: 735 nm)

5

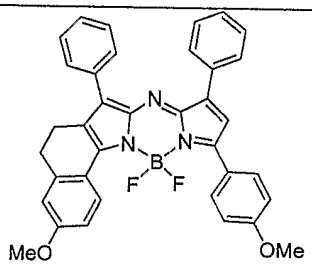
Table 1 (Continued)



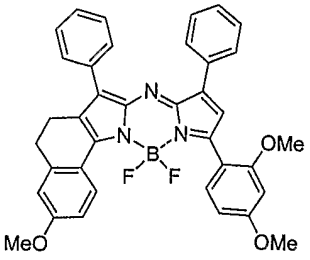
4 (λ_{max}: 688 nm)



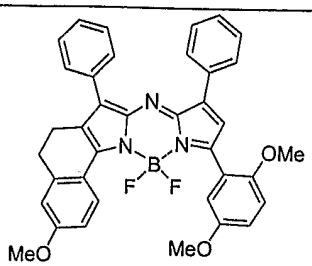
5 (λ_{max}: 678 nm)



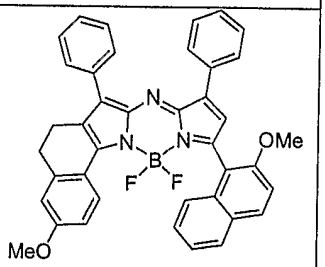
6 (λ_{max}: 708 nm)



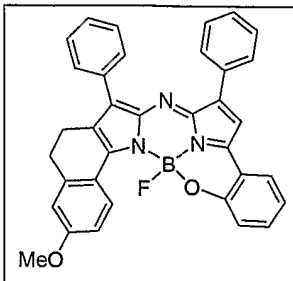
7 (λ_{max}: 707 nm)



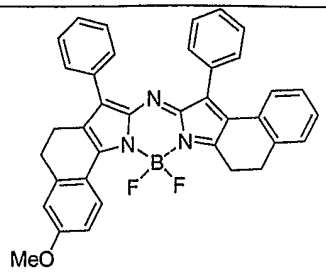
8 (λ_{max}: 681nm)



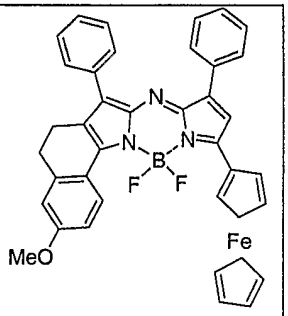
9 (λ_{max}: 665 nm)



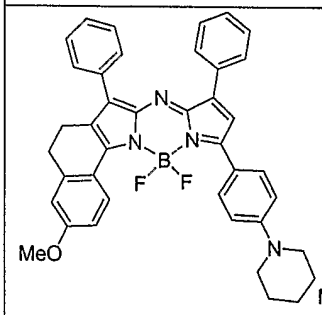
10 (λ_{max}: 735 nm)



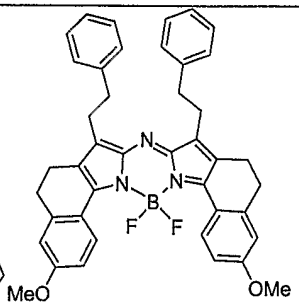
11 (λ_{max}: 714 nm)



12 (λ_{max}: 739 nm)

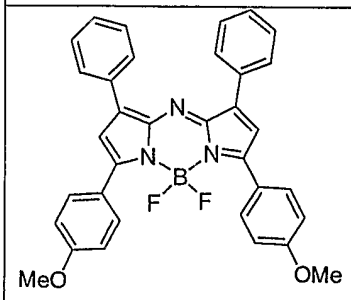


13 (λ_{max}: 778 nm)



14 (λ_{max}: 707 nm)

Table 1 (Continued)

Reference Dye


15
15 λ_{max}: 688 nm

The present invention is illustrated by the following examples, which are in no way intended to limit the scope of the present invention. Numerous modifications and variations will be apparent to those skilled in the art.

Examples

General Procedure for preparation of azadipyrromethene of formula (VII) or (VIII)

Procedure A:

To a mixture of pyrrole derivative (0.2 mmol) in a mixture of acetic acid (1 ml) and acetic anhydride (0.4 ml) was added in one portion of $NaNO_2$ (6.9 mg, 0.1 mmol) with stirring under ice-water cooling. The mixture was stirred for 30 minutes at ambient temperature followed by heating at 80 °C for 10 minutes. The reaction mixture was cooled down, quenched with crushed ice. The resulted solid was collected by filtration, and was used for the next step without further purification.

Procedure B:

Pyrrole derivative (0.1 mmol) was dissolved in acetic acid (1 ml) by heating, the mixture was quickly cooled down in a water bath (about 10° C), $NaNO_2$ (0.1 mmol) was added with stirring, stirred for 10 minutes, warmed to room temperature, followed by addition of acetic anhydride (0.4 ml). The second pyrrole moiety (0.1 mmol) was added in one portion, stirred for 30 minutes at ambient temperature followed by heating at 80 °C for 10 minutes. The reaction mixture was cooled down, quenched with crushed ice, neutralized with aqueous sodium carbonate, and extracted with dichloromethane. The organic solution was filtered through a pad of silica gel, washed with dichloromethane. After removal of solvent, the resulted solid was used for the next step without further purification.

Procedure C:

The Procedure B was followed except that acetic acid (0.5 ml) instead of acetic anhydride was added after nitrosation step.

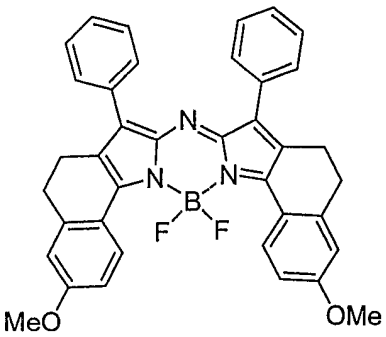
General Procedure for preparation of azadipyrromethene complex

Procedure D:

The solid of crude azadipyrromethene was added 1,2-dichloromethane (3 ml) dried over molecular sieves (3 Å) and Hünig's amine (~5 eq). Boron trifluoride diethyl ether (~4.5 eq) was added dropwise with stirring at room temperature. The mixture was stirred for 5 minutes, followed by heating at 80 °C for 10 minutes. The complex formation reaction was monitored by UV spectroscopy. More Hünig's amine and Boron trifluoride diethyl ether may be required until UV spectra indicated complete conversion. The mixture was cooled down, quenched with water. Organic laser was separated, aqueous layer was extracted with dichloromethane. The combined organic solution was filtrated through a pad of alumina (activity III). Solvent was removed, the residue was digested with dichloromethane/hexane mixture, and recrystalized from dichloromethane/hexane. The mother liquid was purified by chromatography on alumina (activity III) and recrystalized from dichloromethane/hexane.

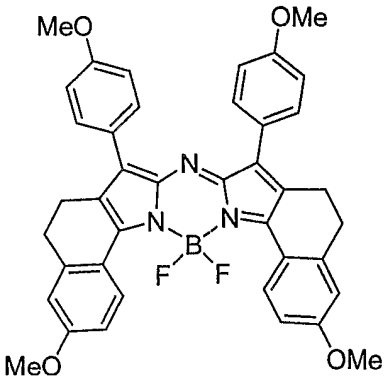
Example 1

Compound **1**



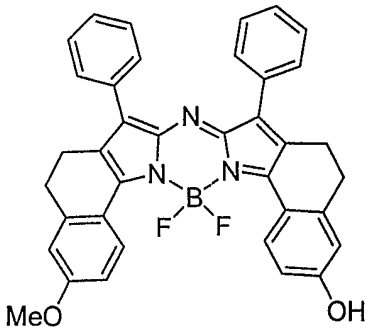
The general procedures A and D were followed starting from 7-methoxy-3-phenyl-4,5-dihydro-1H-Benz[g]indole, 46.6 mg coppery crystal was obtained (76.5%).

Compound **2**



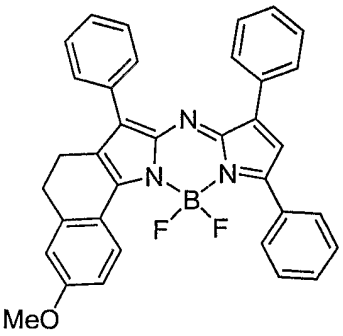
The general procedures A and D were followed starting from 7-methoxy-3-(*p*-methoxyphenyl)-4,5-dihydro-1H-Benz[g]indole, coppery crystal was obtained (80.2%).

Compound **3**



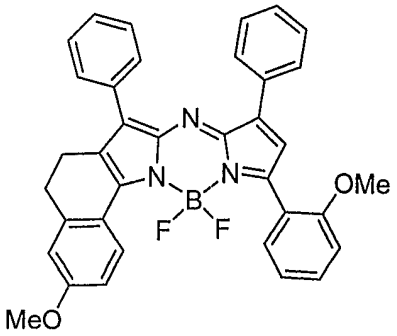
The general procedures C and D were followed, starting from 7-methoxy-3-phenyl-4,5-dihydro-1H-Benz[g]indole, then with 2-(*p*-hydroxyphenyl)-4-phenyl-1H-pyrrole, coppery crystal was obtained.

Compound **4**



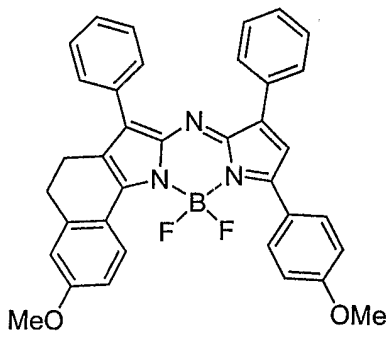
The general procedures B and D were followed, starting from 7-methoxy-3-phenyl-4,5-dihydro-1H-Benz[g]indole, then with 2,4-diphenyl-1H-pyrrole, 28.2 mg coppery crystal was obtained (52%).

Compound **5**



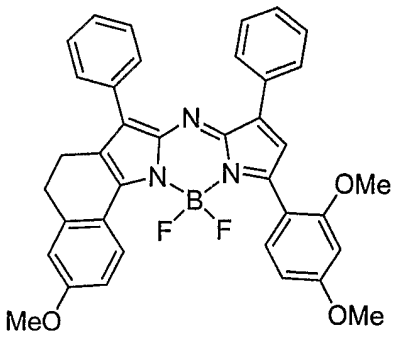
The general procedures B and D were followed, starting from 7-methoxy-3-phenyl-4,5-dihydro-1H-Benz[g]indole, then with 2-(*o*-methoxyphenyl)-4-diphenyl-1H-pyrrole, coppery crystal was obtained.

Compound **6**



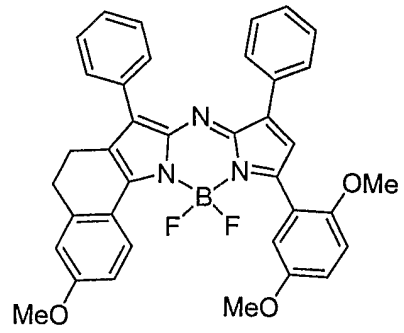
The general procedures B and D were followed, starting from 7-methoxy-3-phenyl-4,5-dihydro-1H-Benz[g]indole, then with 2-(*p*-methoxyphenyl)-4-diphenyl-1H-pyrrole, coppery crystal was obtained.

Compound **7**



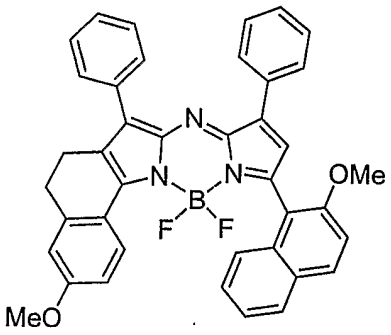
The general procedures B and D were followed, starting from 7-methoxy-3-phenyl-4,5-dihydro-1H-Benz[g]indole, then with 2-(*o*,*p*-dimethoxy-diphenyl)-1H-pyrrole, coppery crystal was obtained.

Compound **8**



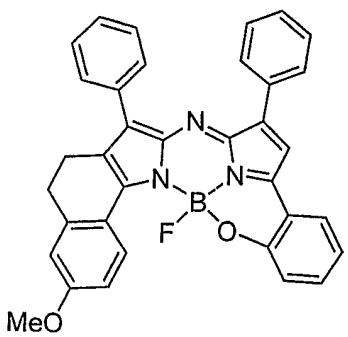
The general procedures B and D were followed, starting from 7-methoxy-3-phenyl-4,5-dihydro-1H-Benz[g]indole, then with 2-(*o*,*m*-dimethoxyphenyl)-4-diphenyl-1H-pyrrole, coppery crystal was obtained.

Compound **9**



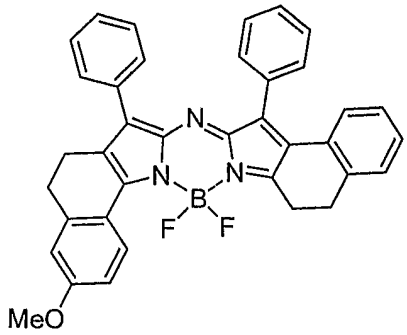
The general procedures B and D were followed, starting from 7-methoxy-3-phenyl-4,5-dihydro-1H-Benz[g]indole, then with 2-(2-methoxy-naphthyl-1-yl)-4-phenyl-1H-pyrrole, coppery crystal was obtained.

Compound **10**



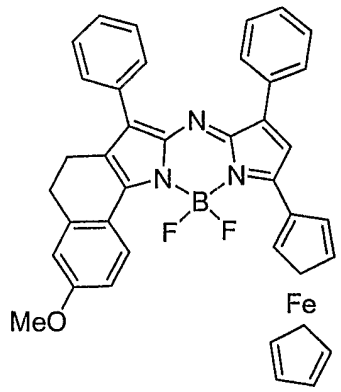
The general procedures C and D were followed, starting from 7-methoxy-3-phenyl-4,5-dihydro-1H-Benz[g]indole, then with 2-(*o*-hydroxyphenyl)-4-phenyl-1H-pyrrole, coppery crystal was obtained.

Compound **11**



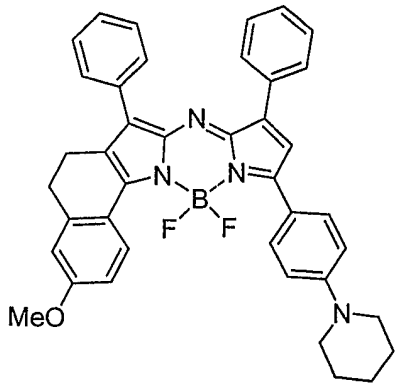
The general procedures B and D were followed, starting from 7-methoxy-3-phenyl-4,5-dihydro-1H-Benz[g]indole, then with 3-phenyl-8,9-dihydro-1H-benzo[e]indole, coppery crystal was obtained.

Compound **12**



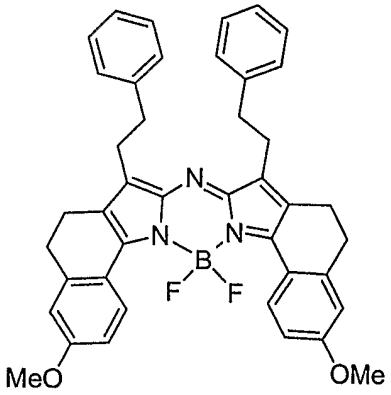
The general procedures B and D were followed, starting from 7-methoxy-3-phenyl-4,5-dihydro-1H-Benz[g]indole, the with 2-ferroceno-2-phenyl-1H-pyrrole, coppery crystal was obtained.

Compound **13**



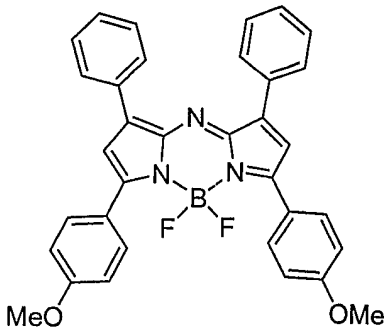
The general procedures B and D were followed, starting from 7-methoxy-3-phenyl-4,5-dihydro-1H-Benz[g]indole, the with 2-(*p*-piperidinophenyl)-4-phenyl-1H-pyrrole, red crystal was obtained.

Compound **14**



The general procedures A and D were followed, starting from 7-methoxy-3-phenylethyl-4,5-dihydro-1H-Benz[g]indole, coppery crystal was obtained.

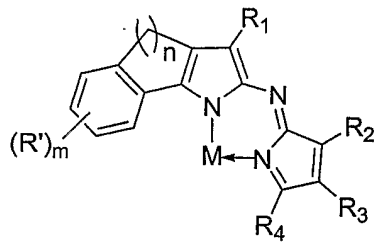
Reference compound **15**



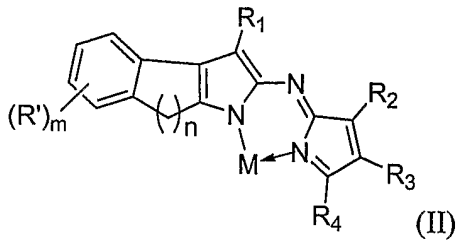
General procedures A and D were followed, starting from 2-(*p*-methoxyphenyl)-4-phenyl-1H-pyrrole, 56 mg coppery crystal was obtained (100%).

Claims

1. Compound of the general formula (I) or (II)



(I)



(II)

or a salt, metal complex or hydrate thereof

wherein

M is a chelating agent select from BF, BF_2, BCl, BCl_2, Zn, Mg, Al, Si, Sn, Cu, Ni, Co.

R' is hydrogen, hydroxy, halogen, nitro, cyano, allyl, linear or branched $(C_1\text{-}C_{20})$alkyl, $(C_3\text{-}C_{20})$cycloalkyl, $(C_1\text{-}C_{20})$alkoxy, $(C_1\text{-}C_{20})$alkylacetylenyl, phenylacetylenyl, $(C_2\text{-}C_{20})$alkenyl, phenylvinyl, halo$(C_1\text{-}C_{20})$alkyl, halo$(C_3\text{-}C_{20})$cycloalkyl, halo$(C_1\text{-}C_{20})$alkoxy, aryl, aryloxy or heteroaryl optionally substituted with $(C_1\text{-}C_6)$alkyl or $(C_1\text{-}C_6)$alkoxy; arylalkyl or heteroarylalkyl; nitrogen-containing heterocycloalkyl having 5 or 6 atoms optionally substituted with $(C_1\text{-}C_6)$alkyl or $(C_1\text{-}C_6)$alkoxy, $-N(R_A)R_B$, $CON(R_A)R_B$, wherein R_A and R_B may be the same or different and are each independently hydrogen, $(C_1\text{-}C_{20})$alkyl, $(C_3\text{-}C_{20})$cycloalkyl, and optionally substituted phenyl; –OCOR, –COOR or –COR, wherein R represents hydrogen, $(C_1\text{-}C_{20})$alkyl, $(C_3\text{-}C_{20})$cycloalkyl, or aryl or heteroaryl optionally

substituted with (C_1-C_6 carboxylate)alkyl or (C_1-C_6)alkoxy; or R' may also be a water solubilizing group such as sulfonate, or quaternary ammonium salt;

R_1-R_4 are the same or different and are each independently:

(a) linear or branched (C_1-C_{12})alkyl, (C_3-C_{12})cycloalkyl, aryl(C_1-C_6)alkyl or heteroaryl(C_1-C_6)alkyl, (C_1-C_6)alkoxy(C_1-C_6)alkyl, (C_1-C_{12})alkoxy, halo(C_1-C_{12})alkyl, (C_1-C_{12})haloalkoxy, (C_1-C_{12})alkylthio;

(d) substituted or unsubstituted aryl groups;

(e) substituted or unsubstituted heteroaryl groups;

(d) a group of the following formulae:

wherein B is hydrogen, (C_1-C_{12})alkyl or substituted or unsubstituted aryl;

(e) unsubstituted or mono-substituted pyrazolyl, pyridyl, imidazolyl, pyrazolinyl, imidazolinyl, or acridinyl, each of the said substituents being (C_1-C_6)alkyl, (C_1-C_6)alkoxy, fluoro, chloro, or phenyl.

(f) a group of the following formulae:

wherein C and D may be the same or different and are each independently carbon, oxygen, (C_1-C_{12})alkyl nitrogen, or (C_1-C_{12})acyl nitrogen;

R_C and R_D are each hydrogen or (C_1-C_{12})alkyl; and

wherein the phenyl moiety is optionally substituted with (C_1-C_{12})alkyl, (C_1-C_{12})alkoxy, (C_2-C_{12})acyl, fluoro, or chloro ;

(g) a water solubilizing group such as sulfonate, carboxylate, or quaternary ammonium salt;

(h) R_2 and R_3, R_3 and R_4 may be joined to form a ring with ring size of 5, 6 or 7;

n is an integer from 1 to 4; and

m is an integer from 0 to 4.

2. Compound of the general formula (I) or (II) according to claim 1, wherein:

M is BF, BF_2, Zn, Mg, Cu, Ni, Co;

R' is selected from hydrogen, nitro, cyano, allyl, fluoro, chloro, bromo, trifluoromethyl, trichloromethyl, dimethylamino, diethylamino, pyrrolidino, piperidino, morpholino, phenyl, benzyl; linear or branched (C_1-C_6)alkyl, (C_1-C_6)alkoxy, or –OCOR or –COOR wherein R is hydrogen, (C_1-C_6)alkyl, (C_3-C_6)cycloalkyl, or R' may also be a water solubilizing group such as sulfonate, carboxylate, or quaternary ammonium salt;

R_1-R_4 are the same or different and are each independently:

(a) linear or branched (C_1-C_6)alkyl, (C_3-C_6)cycloalkyl, aryl(C_1-C_4)alkyl or heteroaryl(C_1-C_4)alkyl, (C_1-C_6)alkoxy(C_1-C_6)alkyl;

(b) unsubstituted, mono-, di-substituted aryl selected from phenyl or naphthyl, preferably substituted in the ortho, the para position, or both;

(c) unsubstituted or mono-substituted heteroaryl groups that are furyl, thienyl, 2,2'-bithienyl, pyrryl, indolyl, benzofuryl, benzothienyl, pyridyl, dibenzofuryl, dibenzothienyl, or carbazolyl the substituents being

amino, cyano, hydroxy, hydroxyethoxy, methoxyethoxy, hydroxyethoxyethoxy, methoxyethoxyethoxy, fluoro, chloro, bromo, iodo, vinyl, allyl, trifluoromethyl, phenyl, (C_1-C_6)alkyl, $(C_1-$

C_6)alkoxy, cyclo(C_3- C_6)alkyl, cyclo(C_1-C_6)alkoxy, (C_1-C_6)alkylamino, di(C_1-C_6)alkylamino, diarylamino, phenylacetylenyl, or phenylvinyl;

a heterocyclic nitrogen-containing substituent, such as N(C_1-C_6)alkylpiperazino, N-aryl-piperizino, aziridino, indolino, pyrrolidino, pyrrolino, piperidino, (C_1-C_4)alkylpiperidino, di(C_1-C_4)alkylpiperidino, 4-piperidinopiperidino, morpholino, 2,6-di(C_1-C_4)alkylmorpholino, thiomorpholino, thioazolidino, tetrahydroquinolino, or pyrryl;

$N(R_A)R_B$, $CON(R_A)R_B$, wherein R_A and R_B are the same or different and are each independently hydrogen, (C_1-C_6)alkyl, (C_3-C_6)cycloalkyl, phenyl or –COR, –OCOR or –COOR wherein R is hydrogen, (C_1-C_6)alkyl, (C_3-C_6)cycloalkyl, or phenyl;

(d) a water solubilizing group such as sulfonate, carboxylate, or quaternary ammonium salt;

(e) R_2 and R_3, R_3 and R_4 may be joined to form a ring with ring size of 5, 6 or 7;

n is an integer from 1 to 3; and

m is an integer from 0 to 2.

3. The compound of the general formula (I) or (II) according to claim 1, wherein:

M is BF or BF_2;

R' is hydrogen, fluoro, chloro, bromo, dimethylamino, diethylamino, pyrrolidino, piperidino, morpholino, phenyl, benzyl, (C_1-C_4)alkyl, or (C_1-C_4)alkoxy, or R' may also be a water solubilizing group such as sulfonate, carboxylate, or quaternary ammonium salt;

R_1-R_4 are the same or different and are each independently:

unsubstituted, mono-, or di-substituted phenyl, preferably substituted in the alpha position, para position or both with the substituents being one or more of amino, acyl, cyano, methoxy, ethoxy, methoxyethoxy, fluoro, chloro, vinyl, allyl, methoxycarbonyl, ethoxycarbonyl, (C_1-C_4)alkyl, di(C_1-C_4)alkylamino, piperazino, piperidino, arylperidino, morpholino, pyrrolidino, aziridino, acryloxy, methacryloxy, phenylacetylenyl, phenylvinyl;

unsubstituted, mono-substituted heteroaromatic groups, such as furyl, thienyl, 2,2'-bithienyl, pyrryl, substituted with a substituent that is (C_1-C_4)alkyl or phenyl;

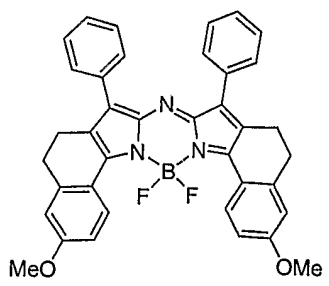
a water solubilizing group such as sulfonate, carboxylate, or quaternary ammonium salt;

R_2 and R_3, R_3 and R_4 may be joined to form a ring with ring size of 5 or 6;

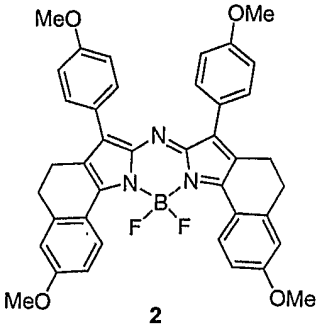
n is an integer from 1 to 3, and

m is, independently, integer from 0 to 2.

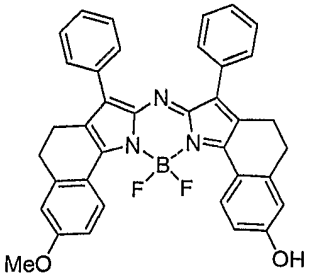
4. A compound of the general formula (I) or (II) having any of the following structures:



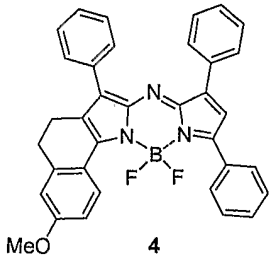
1



2



3



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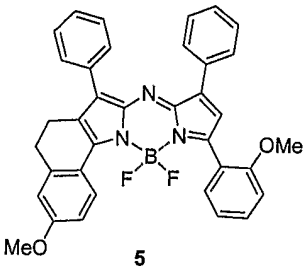

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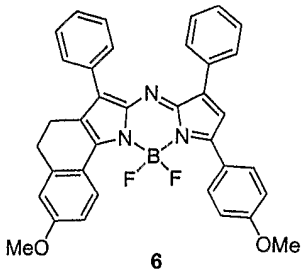

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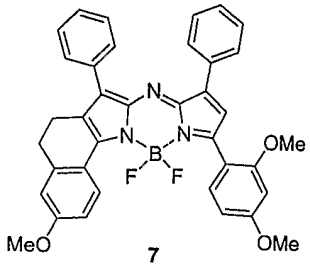

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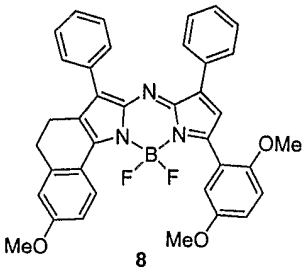

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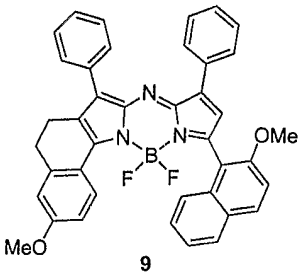

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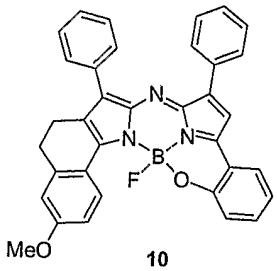

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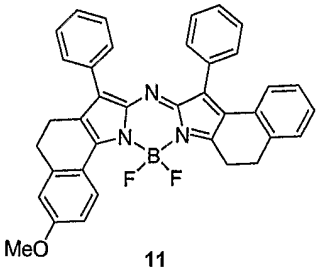

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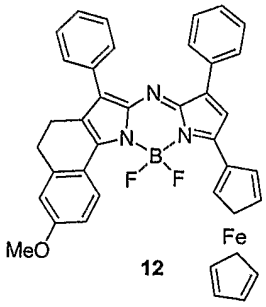

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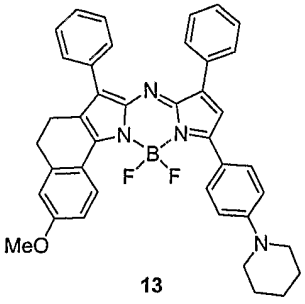

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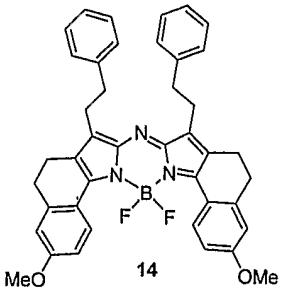

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5. A compound according to anyone of claims 1 to 4 for use in bio sensing, fluorescent labeling, in optical recording or in photodynamic therapy in vivo or in vitro.

6. A method for producing a compound according to claims 1 to 4 comprising the step of in situ preparation of a nitrosopyrrole intermediate followed by complex formation to arrive at the compounds of the invention.

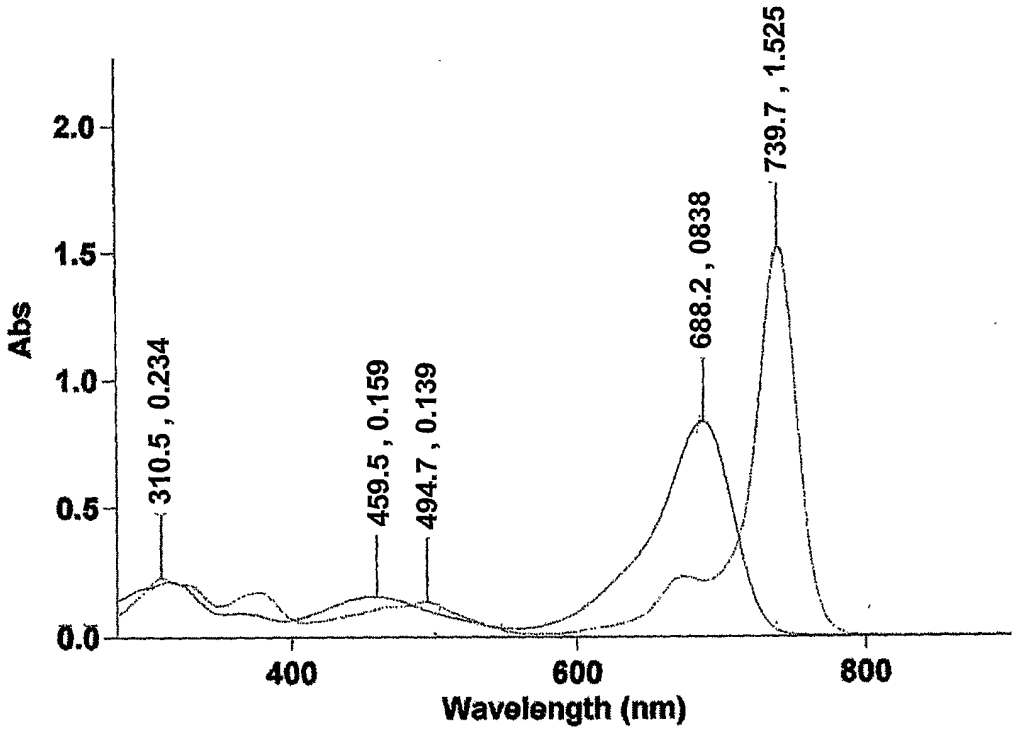

Fig.1. Absorption spectra of compound **1** and reference compound **15** (1.0×10^{-5} mol/l) in chloroform.

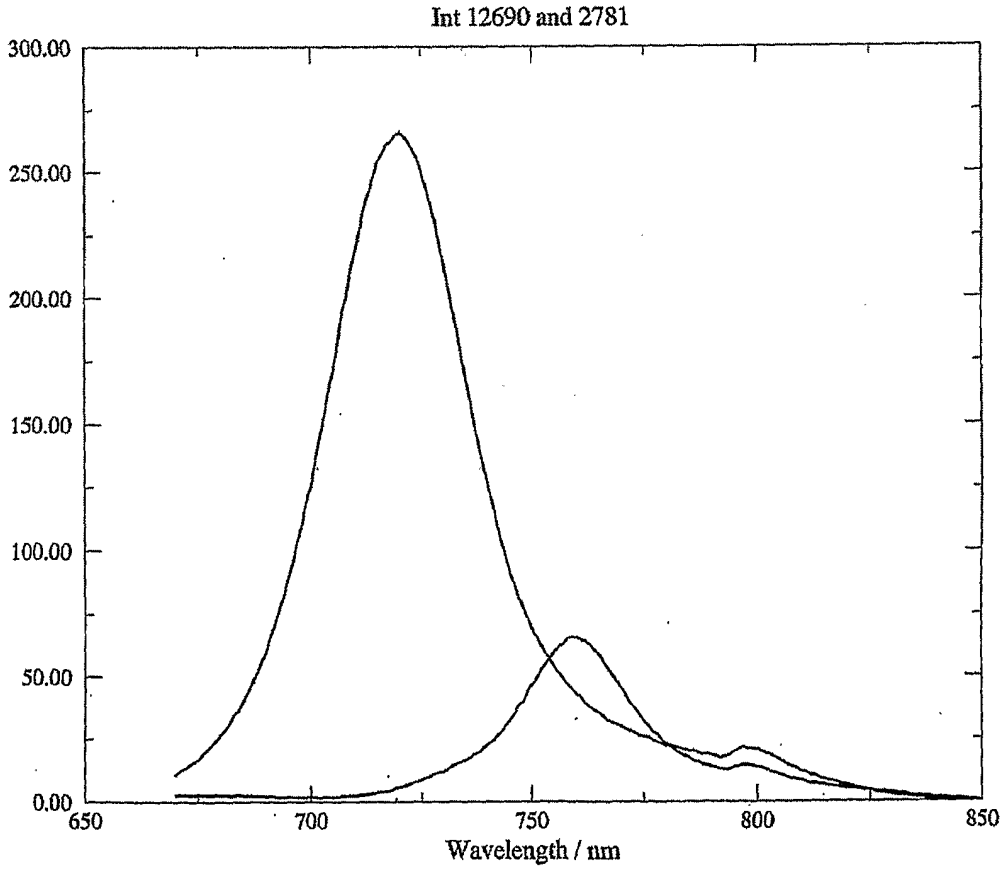

Fig.2. Fluorescent spectra of compound **1** and reference compound **15** excited at 650 nm under identical condition at room temperature.

INTERNATIONAL SEARCH REPORT

International application No
PCT/CH2005/000709

A. CLASSIFICATION OF SUBJECT MATTER
C09B55/00 G11B7/246

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)
C09B

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, CHEM ABS Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	WO 03/080627 A (UNIVERSITY COLLEGE DUBLIN, NATIONAL UNIVERSITY OF; O'SHEA, DONAL; KILL) 2 October 2003 (2003-10-02) cited in the application compound 2B	1-5
Y	A.GORMAN ET AL.: "In vitro Demonstration of the Heavy-Atom Effect for Photodynamic Therapy" J.AM.CHEM.SOC., vol. 126, 2004, pages 10619-10631, XP002374818 Conclusion and compounds 18 A-D	1-6

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

[X] See patent family annex.

* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

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"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

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"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

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"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
29 March 2006	19/04/2006
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 Friebel, F

INTERNATIONAL SEARCH REPORT

International application No
PCT/CH2005/000709

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	J.CHEN ET AL.: "4,4-Difluoro-4-bora-3a,4a-diaza-s-indacene (BODIPY) Dyes modified for extended Conjugation and restricted Bond Rotations" J.ORG.CHEM., vol. 65, 2000, pages 2900-2906, XP002374819 Conclusion and compound 4 -----	1-5
P,X	W.ZHAO ET AL.: "Conformationally restricted Aza-Bodipy: A Highly Fluorescent, stable, Near-Infrared-Absorbing Dye" ANGEW.CHEM.INT.ED., vol. 44, 2005, pages 1677-1679, XP002374820 the whole document -----	1-6
Y	US 2 469 830 A (KNOTT EDWARD BOWES) 10 May 1949 (1949-05-10) cited in the application the whole document -----	6

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No
PCT/CH2005/000709

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 03080627 A	02-10-2003	AU 2003224000 A1	08-10-2003
		EP 1492799 A1	05-01-2005
		US 2005107335 A1	19-05-2005
US 2469830 A	10-05-1949	BE 477023 A	