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Synthesis and Characterization of Glycoconjugated Porphyrin Triphenylamine Hybrids for Targeted Two-Photon Photodynamic Therapy

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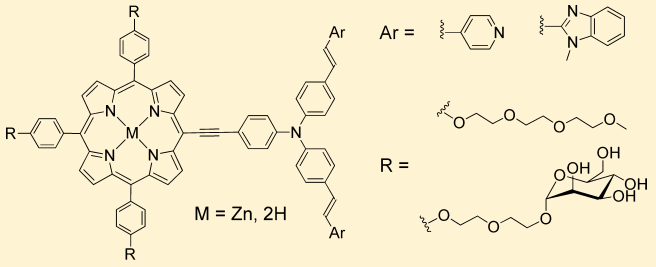
Supporting Information

ABSTRACT: In order to avoid side effects at the time of cancer eradication to the patients, the selectivity of treatments has become of strategic importance. In the case of photodynamic therapy (PDT), two-photon absorption combined with active targeting of tumors could allow both spatial and chemical selectivity. In this context, we present the synthesis, spectroscopic, and biological properties of a series of porphyrin-triphenylamine hybrids with excellent singlet oxygen production capacities and good two-photon absorption.



INTRODUCTION

Photodynamic therapy (PDT) is an emerging treatment designed to eliminate diseased tissues such as malignant tumors.[1−4] It involves a photosensitizing molecule (PS) that can be activated to its triplet excited state and transfers its energy to molecular triplet oxygen resulting in the generation of singlet oxygen or other reactive oxygen species (ROS). These highly reactive species lead to the destruction of targeted diseased tissues by biochemical damage. A major advantage of PDT in comparison with other treatments such as chemo- or radiotherapy is that, in the absence of light activation, the PS is well-tolerated by the cells without cytotoxic effect. Therefore, PDT has the potential to selectively destroy malignant cells while sparing the nonirradiated normal tissues.

PS currently used in PDT are mainly cyclic tetrapyrroles or porphynoids because of their long excited triplet-state lifetime.[5] These PS in free-base form have five one-photon absorption (1PA) bands in the visible wavelength range (400−700 nm), and despite promising results, an important limitation arises from the fact that the penetration depth of these wavelengths is confined near the surface of the tissues. However, the absorption is much lower in the optical window of biological tissues between 700 and 1300 nm to obtain an effective photobiological effect.[6] One way to overcome this problem consists of taking advantage of two-photon absorption (2PA) processes. In this nonlinear optical phenomenon, two photons of lower energy are absorbed simultaneously. 2PA−PDT should allow also greater precision than is achievable by conventional one-photon excitation as a consequence of the quadratic dependence of two-photon excitation on the local light intensity.[7] Simultaneous two-photon excitation of porphyrin derivatives can be carried out within the biological optical window. Because classical porphyrin photosensitizers exhibit small 2PA values using femtosecond pulsed lasers (less than 50 GM where 1 GM = 10^{-50} $cm^4 \cdot s \cdot molecules^{-1}$),[8,9] molecular engineering is necessary to improve the 2PA properties unless it becomes necessary to use very high laser intensities that can be deleterious for the biological tissues.[10,11] The directions that have been explored consist of more extended cyclic polypyrroles,[12,13] dimers, or oligomers of conjugated porphyrins[14−18] or the use of 2PA light-harvesting antenna.[19−21] Spangler and co-workers have developed porphyrin−triphenylamine hybrids for 2PA−PDT.[22−24] In these structures, a cooperative effect has been demonstrated: these compounds exhibit interesting 2PA cross sections superior to the sum of the cross sections of the two parts taken separately. Spangler showed that, in these compounds, π-conjugation between the porphyrin and the triphenylamine part is necessary to obtain interesting 2PA properties. Moreover, these structures possess a high singlet oxygen quantum yield.

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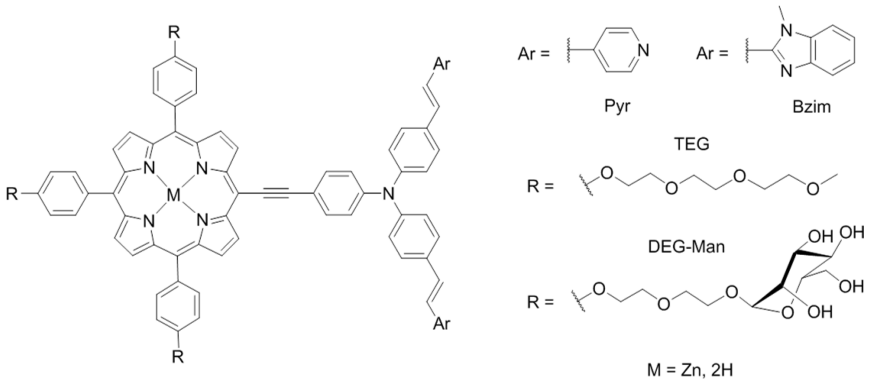


R	M	Ar	Compound
H	Zn	Pyr	1
H	2H	Pyr	2
H	Zn	Bzim	3
TEGOMe	Zn	Pyr	4
DEG-Man	Zn	Pyr	5

Figure 1. Structure of target compounds.

Another limitation of PDT is the low selectivity and specificity of the PS for tumor cell: high drug and light doses are thus required to compensate forthis, leading to damage of healthy tissues. Active targeting of membrane receptors represents an obvious improvement.[25−28] It has been reported that lectin-type receptors are overexpressed in certain malignant cells[29−32] and that carbohydrates such as α-mannose and β-galactose have specific interaction with these receptors.[33−35] Over the past decade, we have focused our efforts on the preparation and in vitro and in vivo evaluation of the 1PA phototoxicity of glycoconjugated tetrapyrrolic macrocycles.[36] Recently, our group has developed a series of glycoconjugated vectorized porphyrin oligomers for 2PA PDT.[37−39]

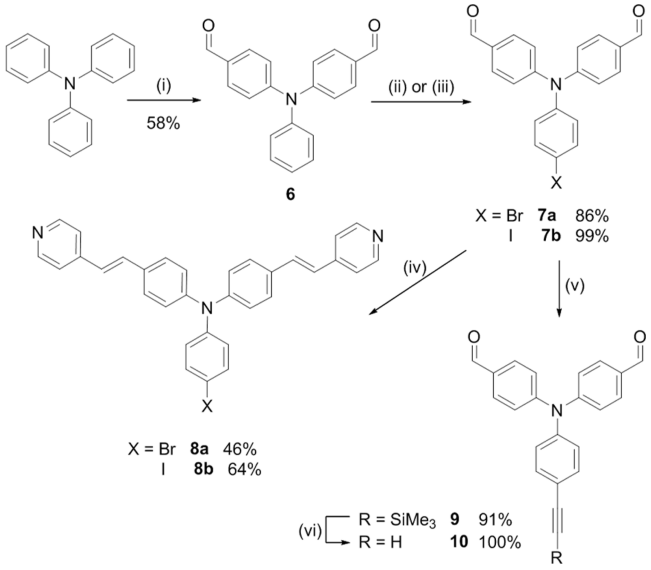
Following the strategy used by Spangler, we have developed a new design of porphyrin−triphenylamine photosensitizer for 2PA PDT exploiting a branching effect. The aim of this work is to describe the synthesis, photophysical (1- and 2PA), and photobiological properties (1PA) of a series of carbohydrate-vectorized porphyrin−triphenylamine hybrids **1**−**5** (Figure 1).

RESULTS AND DISCUSSION

Synthesis. Triphenylamine was converted into the diacylated compound **6** according to a previously described Vilsmeier−Haack procedure using $POCl_3$ and DMF with 58% yield (Scheme 1).[40] The third phenyl ring was halogenated by action of bromine leading to compound **7a** (86% yield)[41] or with iodine and silver sulfate to afford **7b** in quantitative yield. Horner−Wadsworth−Emmons (HWE) reaction of **7a** with (4-pyridinyl)methyl diethyl phosphonate [$PyrCH_2PO(OEt)_2$] led to compound **8a** with trans-geometry (see the NMR results: coupling constant $^3J_{H-H'}$ = 16 Hz characteristic of E configuration, 46% yield) according to the procedure previously developed in our group.[42]

Sonogashira cross-coupling of iodo derivative **7b** with trimethylsilylacetylene (TMSA) gave silylated compound **9** (91% yield), which was deprotected quantitatively by action of tetrabutylammonium fluoride (TBAF), leading to compound **10**. It should be noted that Sonogashira coupling of bromo derivative **7a** was not successful.

Scheme 1. Synthesis of Triphenylamine Residues[a]



[a]Key: (i) DMF, $POCl_3$, 95 °C, 4 h; (ii) Br_2, CH_2Cl_2, reflux, 4 h; (iii) I_2, Ag_2SO_4, EtOH, rt, 15 h; (iv) $PyrCH_2PO(OEt)_2$, NaH, THF, rt, 48 h; (v) X = I, TMSA, $Pd(PPh_3)_2Cl_2$, CuI, THF, Et_3N, rt, 15 h; (vi) TBAF, CH_2Cl_2, 0 °C, 30 min.

5-Bromo-10,15,20-triphenylporphyrinatozinc(II) obtained according to Senge's strategy[43] was converted to silylated compound **11** via Sonogashira coupling in 89% yield. The synthesis of the target compounds **1**−**3** via Sonogashira cross-coupling was first considered (Scheme 2), taking into account the fact that such cross coupling of 5,10,15-triphenyl-20-(trimethylsilyl)ethynylporphyrinatozinc(II) **11** has previously been described.[1,8] However, reaction of **11** with triphenylamine derivative **8a** led to the homocoupling product and desired compound **1** in very small yield (19%) (Scheme 2). An

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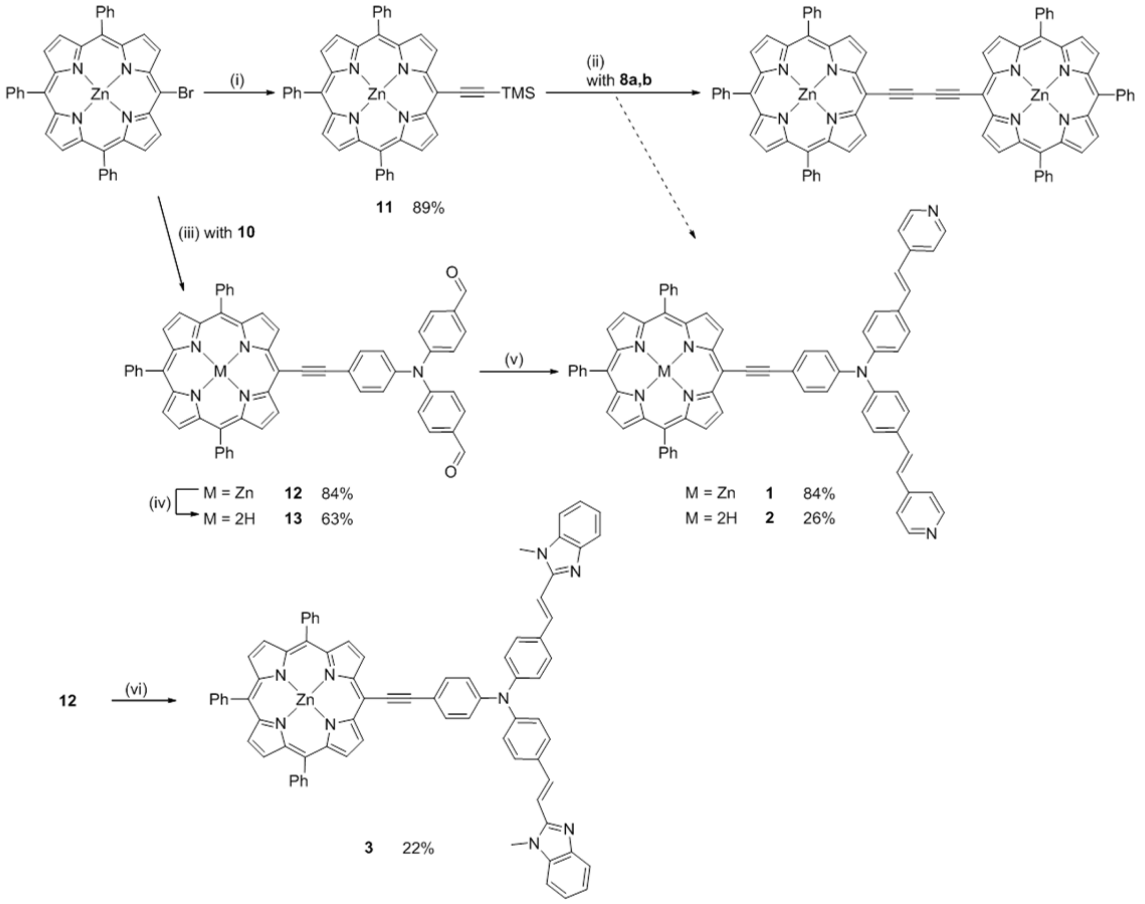
Scheme 2. Synthesis of Compounds 1−3[a]



[a]Key: (i) TMSA, $Pd(PPh_3)_2Cl_2$, CuI, THF, Et_3N, rt, 15 h; (ii) TBAF, CH_2Cl_2, 0 °C, 30 min then $Pd_2(dba)_3$, $AsPh_3$, THF, Et_3N, reflux, 18 h; (iii) **10**, $Pd(PPh_3)_2Cl_2$, CuI, THF, Et_3N, rt, 20 h; (iv) TFA, $CHCl_3$; (v) $PyrCH_2PO(OEt)_2$, NaH, THF, rt, 48 h; (vi) $BzimCH_2PPh_3Cl$, DBU, THF, rt, 15 h.

alternative strategy was thus adopted, involving compound **12** which was obtained from **10** and 5-bromo-10,15,20-trisphenylporphyrinatozinc(II) by Sonogashira coupling reaction (84% yield). The zinc atom was removed by action of trifluoroacetic acid (TFA), leading to the free base analogue **13** (63% yield). Compounds **12** and **13** were then converted to target compounds **1** and **2** by HWE reaction (with yields of 84 and 26%, respectively, exclusively in *E* configuration as was confirmed by ^{1}H NMR with a coupling constant $^3J_{H-H'}$ of about 16 Hz for the alkenyl hydrogens). Wittig reaction between **12** and (*N*-methylbenzimidazolyl)methyltriphenylphosphonium chloride ($BzimCH_2PPh_3Cl$) in the presence of 1,8-diazabicycloundec-7-ene (DBU) afforded the compound **3** in 22% yield.

The second-generation compound **4** decorated with water-solubilizing TEG fragments has also been designed (Scheme 3). 5,10,15-Tris(4-hydroxyphenyl)porphyrin **15** was obtained by the deprotection of isopropoxy groups with BBr_3 of 5,10,15-tris(4-isopropyloxyphenyl)porphyrin.[43,44] Bromination with *N*-bromosuccinimide (NBS) (compound **16**) and metalation with zinc(II) acetate afforded compound **17** with 81% yield over two steps. Then, 2-[2-(2-methoxyethoy)ethoxy]ethanol tosylate **14** and **17** reacted through a Williamson coupling reaction leading to **18** with 88% yield. Sonogashira cross-coupling reaction with TMSA afforded compound **19** with 69% yield. Compound **19** was deprotected with TBAF and coupled with **7b** to afford **20** (95% yield), which was engaged in the HWE reaction with $PyrCH_2PO(OEt)_2$ giving compound **4** with 99% yield (*E* geometry: see NMR results, characteristic coupling constant $^3J_{H-H'} \approx 16$ Hz).

The synthesis of a third-generation compound bearing α-mannose targeting moieties is described in Scheme 4. The Williamson's reaction between **15** and 2-bromoethoxyethoxy-*O*-2′,3′,4′,6′-tetraacetyl-α-D-mannose, previously described by our team,[45] led to compound **21** which is converted in three steps in **22**.[36] Unfortunately, the Sonogashira cross-coupling reaction[46] does not afford the desired diacyl intermediary **25** and could not be optimized.

Another synthetic pathway was tested. Direct *meso*-functionalization of **15** led to **23** (48% over three steps), whose Sonogashira coupling with **7b** was efficient leading to compound **24** with a 69% yield. The glycosyl moieties were added on compound **24** by Williamson's reaction to afford compound **25** (49%). Unfortunately, the pyridinyl groups could not be introduced on **25** to obtain compound **30**. The same synthetic problems were encountered to convert compound **24** in substrate **29** due to the deprotonation of the phenol groups inducing the precipitation of the substrate.

A third synthetic strategy was envisaged: Tris-isopropoxyphenylporphyrin was brominated on its free *meso* position, metalated with zinc salt, and coupled to TMSA to afford compound **26** in a yield of 78% over three steps. Sonogashira coupling reaction with **7b** led to **27** (19%), and a HWE reaction, directly followed by the deprotection of the alkoxy groups, afforded **29** as a free base analogue (12% over two

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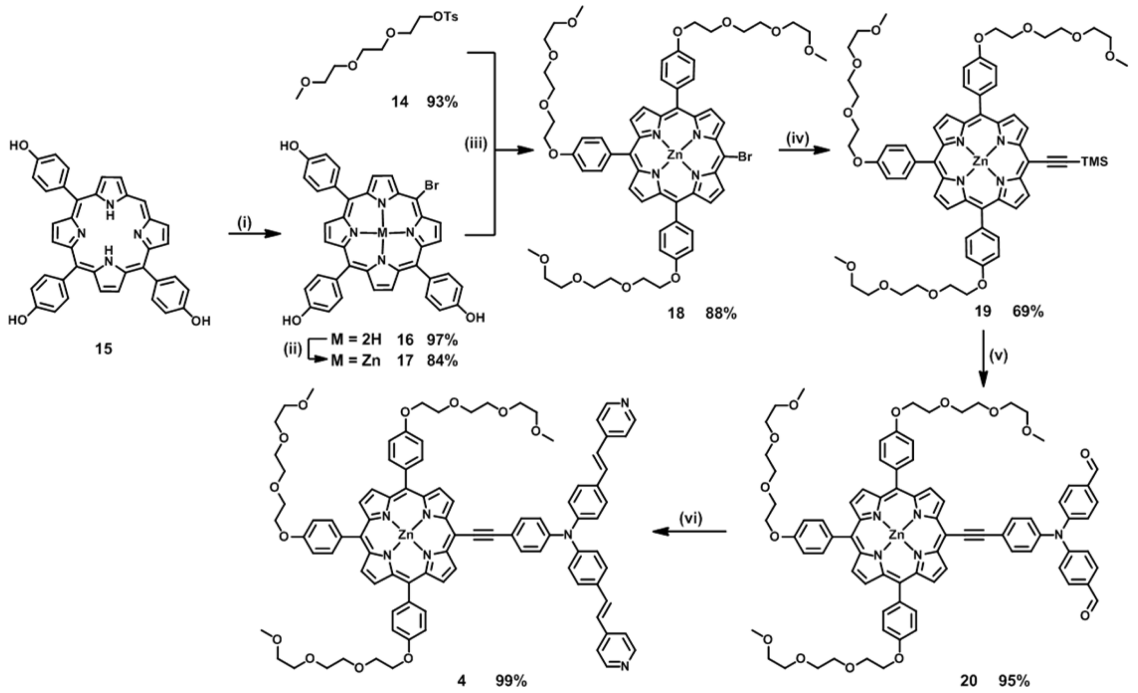
Scheme 3. Synthesis of Compound 4[a]



[a]Key: (i) NBS, $CHCl_3$, pyridine, 0 °C, 30 min; (ii) $Zn(OAc)_2$, $CHCl_3$, MeOH, reflux, 5 min; (iii) Cs_2CO_3, DMF, rt, 18 h; (iv) TMSA, $Pd(PPh_3)_2Cl_2$, CuI, THF, Et_3N, rt, 4 h; (v) TBAF, CH_2Cl_2, 0 °C, 30 min then **7b**, $Pd_2(dba)_3$, $AsPh_3$, THF, Et_3N, rt, 20 h; (vi) $PyrCH_2PO(OEt)_2$, NaH, THF, rt, 48 h.

steps, *E* configuration). Compound **29** reacted with the bromodiethylene glycol protected mannosyl compound to afford **30** (20%). Compound **30** was metalated under the usual conditions, and its glycosyl moieties were deprotected by Zemplén's method[47] in quantitative yields to afford compound **5**. No anisotropic effect of the macrocycle could be observed on the ^{1}H NMR spectrum for the mannosyl hydrogens signals, proving that the carbohydrate moieties are located close to the equatorial plane of macrocyle. All final compounds were isolated in their *E* configuration as observed by ^{1}H NMR ($^3J_{H-H'} \approx 16$ Hz).

Photophysical Properties. The photophysical properties of compounds **1**−**5** are presented in Table 1. Absorption spectra of compounds **1**−**3** recorded in DMSO (Figure 2) are typical of π-conjugated porphyrins[23] with an intense Soret band between 435 and 448 nm. The zinc complexes **1** and **3** exhibit two Q bands at 580−584 and 634 nm, the latter one being more intense while the free-base analogue **2** shows three Q bands (521, 580, 669 nm). Compound **2** shows an additional band at 376 nm which corresponds to the absorption of the triphenylamine group (TP band). TP absorption is also observed at the same wavelength for **1** and **3** but only as a sideband. Compounds **4** and **5** show similar features in DMF.

Excitation of compounds **1**−**3** in CH_2Cl_2 at 450 nm (Soret band) led to fluorescence emission above 600 nm characteristic of porphyrin with two vibrational bands (Figure 3). Excitation of the triphenylamine part at 380 nm led to the apparition of an additional broad emission centered at 480 nm which was attributed to the fluorescence of the triphenylamine part. Substituted triphenylamines are known for being strongly fluorescent molecules,[48] the weak emission of the triphenylamine part in the hybrids might result from an internal energy transfer from the triphenylamine to the porphyrin. This hypothesis is illustrated in Figure 4, which presents emission spectra of **1**, **8b**, and **12** at the same concentration following excitation at 380 nm. Almost total quenching of the triphenylamine is observed in the hybrid as well as increased porphyrin emission in the hybrid. Similar results (absorption and emission) were obtained for compounds **4** and **5** (see the Supporting Information). All compounds display relatively weak fluorescence emission but excellent photosensitizing properties (Φ_Δ 0.64 to 0.84) in a polar solvent like DMF.

Characterization of the two-photon absorption properties of compounds **1**−**5** was obtained after two-photon-induced fluorescence (TPIF) measurements, with a femtosecond Ti:sapphire laser source delivering 100 fs pulses at 76 MHz repetition rate over the spectral range from 770 to 900 nm. The TPIF intensities of the samples were measured relative to a solution of fluorescein, the ratio of the fluorescent signals enabling further determination of the 2PA cross section (δ) assuming equal one- and two-photon fluorescence quantum yields.[49] The dependence of the TPIF intensity with the power of the incident exciting IR beam was systematically checked, a quadratic dependence being observed only for exciting wavelengths above 830 nm. Oppositely, a linear variation of the fluorescence signal intensity was observed for exciting wavelength below this value, pointing to a one-photon excited fluorescence process. The latter was attributed to hot-band absorption as previously reported for porphyrin oligomers or hybrids.[50] The 2PA cross-section of every compound was thus determined at λ_{exc} = 830 nm; this wavelength led to optimized two-photon fluorescence intensities over the range 830−870 nm under which one-photon hot band absorption phenomena were absent. All hybrids exhibit quite good TPA properties, with two-photon absorption cross section values ranging between 159 and 527 GM. Two-photon absorption appears

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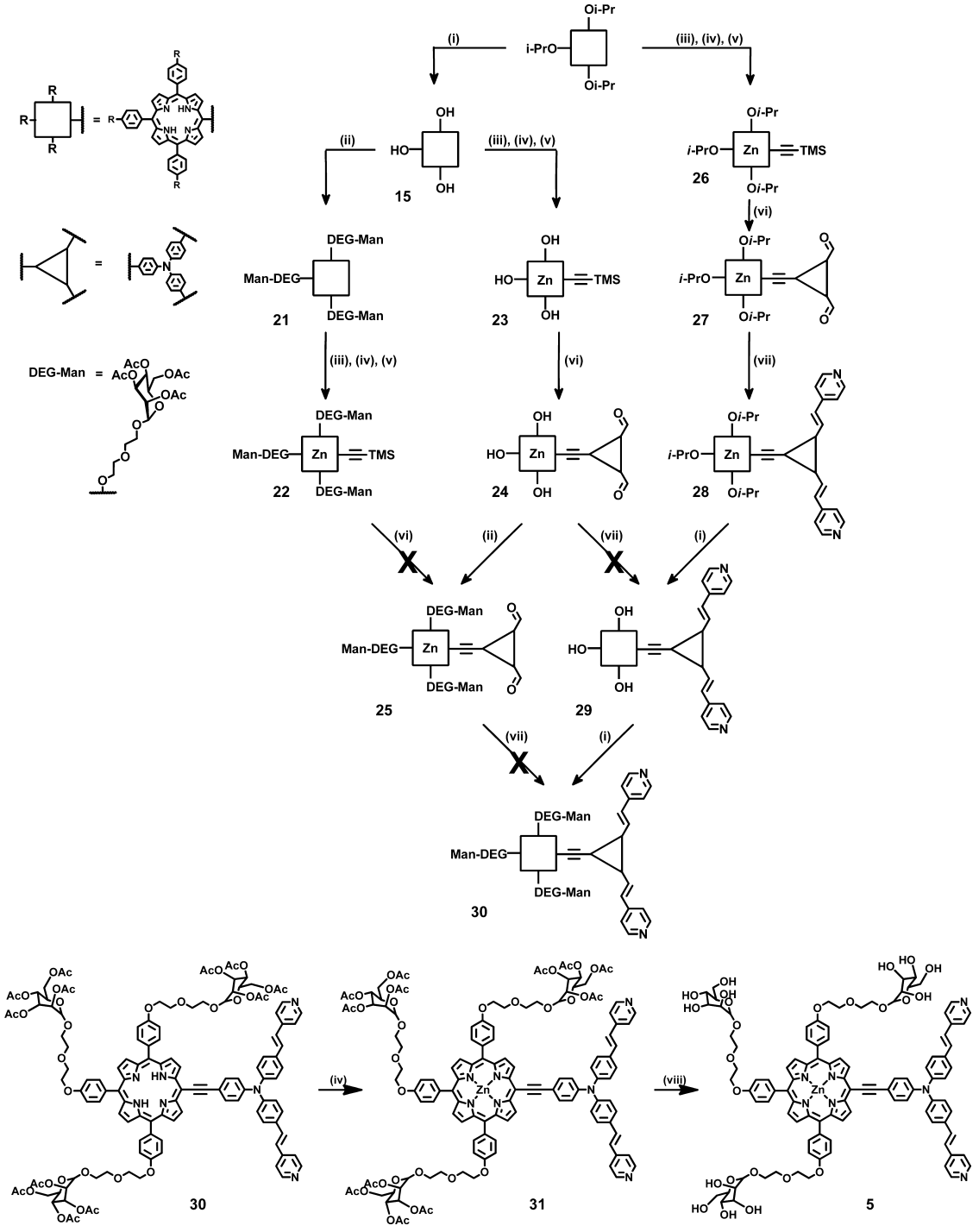
Scheme 4. Synthesis of Compound 5[a]



[a]Key: (i) BBr_3, CH_2Cl_2, −20 °C, 20 min; (ii) 2-[2-(2-bromoethoxy)ethoxy]-*O*-2′,3′,4′,6′-tetraacetyl-α-D-mannose, Cs_2CO_3, DMF, rt, 24 h; (iii) NBS, $CHCl_3$, pyridine, 0 °C, 15 min; (iv) $Zn(OAc)_2$, $CHCl_3$, MeOH, reflux, 5 min; (v) TMSA, $Pd(PPh_3)_2Cl_2$, CuI, THF, Et_3N, rt, 4 h; (vi) TBAF, CH_2Cl_2, 0 °C, 30 min then **7b**, $Pd_2(dba)_3$, $AsPh_3$, THF, Et_3N, rt, 20 h; (vii) $PyrCH_2PO(OEt)_2$, NaH, THF, rt, 48 h; (viii) NaOMe/MeOH, THF, rt, 1 h.

slightly better for compound **1** than for compound **2**, which points at the role played by the zinc atom in the 2PA mechanism. Benzimidazolyl moieties (compound **3**) led to a higher δ_{max} value than pyridyl (compound **2**), probably due to higher 2PA properties of Bzim compounds compared to pyridiniums as was previously reported.[51] It is noteworthy that the modification on the *meso* positions of the porphyrin did not significantly impact the 2PA properties (compounds **1**, **4**, and **5**).

Photobiological Properties. Phototoxicity studies were conducted on two tumor cell lines: Y79 is a commercial line of human retinoblastoma cells overexpressing α-mannose specific lectins,[31] while HT-29 (human colon tumor cells) do not. Only the biologically optimized compounds **4** and **5** were tested.

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Table 1. Photophysical Parameters of Porphyrin Derivatives 1−5[a]

	λ_{abs} (nm)	ε (mM^{-1} cm^{-1})	λ_{em} (nm)	Φ_F	Φ_Δ	δ_{max} (GM)
1	448	160.0	649			
	580	10.6		0.08	0.80	251
	634	30.7				
2	376	103.5				
	435	210.0	678	0.06	0.64	159
	521	20.5	743			
	580	33.2				
	669	15.8				
3	380	75.9				
	446	359.3	649	0.09		527
	584	30.1				
	634	82.6				
4	451	228.8	643			
	582	12.9	708	0.03	0.74	317
	632	40.3				
5	450	159.2	643			
	581	5.8	708	0.02	0.84	225
	639	29.8				

[a] λ_{abs} = maximun absorption peak wavelength in DMSO, ε = molar extinction coefficient. λ_{em} = emission peak in DMF with excitation in Soret band, Φ_F = fluorescence emission quantum yield in DMSO vs coumarin 153 in EtOH (Φ_F = 0.38), Φ_Δ = singlet oxygen quantum yield in DMF vs rose bengal (Φ_Δ = 0.47), δ: 2PA cross-section at 830 nm in DMSO.

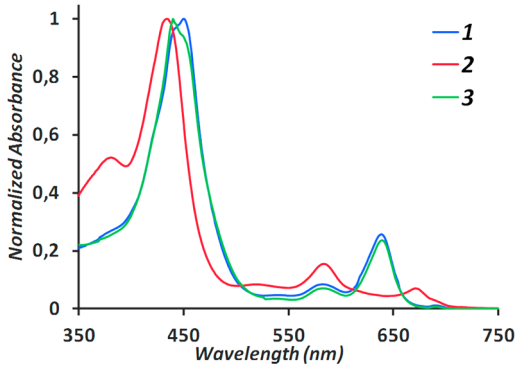

Figure 2. Normalized absorption spectra of compounds **1**−**3** in DMSO.

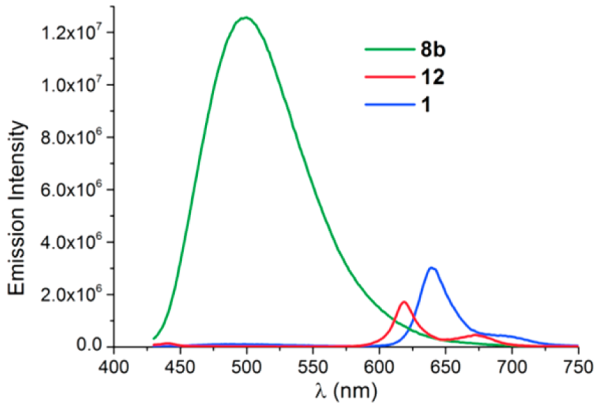

Figure 4. Emission spectra of compounds **1**, **8b**, and **12** at 1 μM in CH_2Cl_2. Excitation at 380 nm.

These tests showed that none of the hybrids displayed any toxicity (dark or light, Table 2) on any cell line. These results were surprising given the high Φ_Δ values of both compounds. Furthermore, they reveal that the active targeting of the membrane receptors with carbohydrates is inefficient. Both observations could be linked to a poor internalization of the hybrids in the cells.

Table 2. Dark Toxicity and Phototoxicity (with Light Illumination: 5 J/cm^2 and Excitation between 400 and 800 nm) of 4 and 5 in Y79 and HT-29 Cell Lines

	dark toxicity IC_{50} (μM)		photocytotoxicity IC_{50} (μM)	
compd	Y79	HT-29	Y79	HT-29
4	>7.5	>7.5	>7.5	>7.5
5	>7.5	>7.5	>7.5	>7.5

The uptake of compounds **4** and **5** in Y79 and HT-29 cells was evaluated by flow cytometry. Two excitation wavelengths were used at 488 nm (foot of the Soret band) and 635 nm (Q-band), and fluorescence emission was monitored above 670 nm in the former case and between 653 and 669 nm for the latter. Although these spectral characteristics were not optimal for PTP compounds, we observed that both hybrids led to partial staining of the cells of both lines with weak fluorescence intensity. The case of **4** on HT-29 showed the highest staining (81−88%). The spectroscopic differences between the two compounds (ε, Φ_F) prevent further comparison.

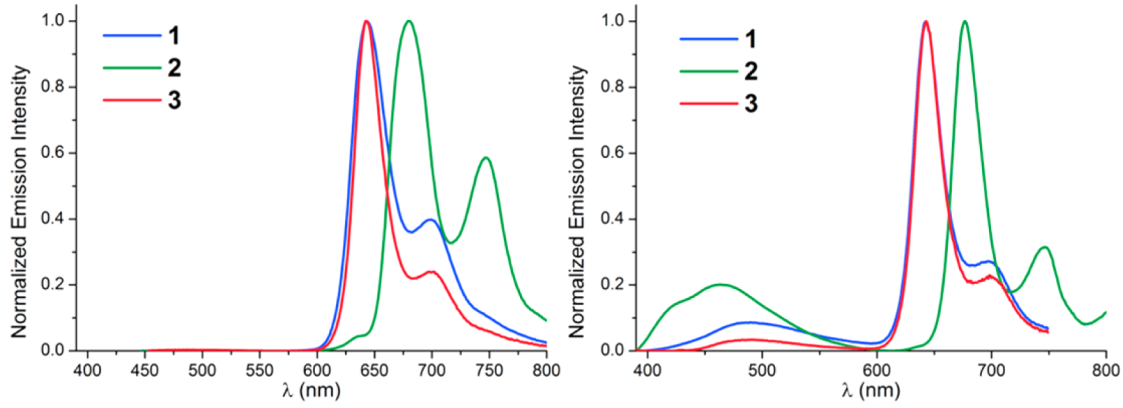

Figure 3. Normalized emission spectra of compounds **1**−**3** in CH_2Cl_2. Excitation at 450 nm (left) and 380 nm (right).

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Table 3. Uptake of Compounds 4 and 5 in Y79 and HT-29 Cell Lines, Compound Detection with the FACSCalibur Characteristics, Cell Percentage of 4 or 5 Positive Cells, and MFI (Treated/Controls Cells) of Fluorescence Intensity

	excitation	488 nm		635 nm	
	emission	FL3-H > 670 nm		FL4-H 653-669 nm	
	Compound	% cell pos	MFI	% cell pos	MFI
Y79	control	0.51	1.00	0.24	1.00
	4	67.10	10.41	47.60	7.95
	5	38.93	8.64	33.43	9.45
HT-29	control	2.47	1.00	2.35	1.00
	4	87.83	30.80	80.83	17.13
	5	50.77	8.95	44.13	7.64

The lack of phototoxicity of the hybrids can therefore be explained by a poor internalization combined with inappropriate intracellular localization or no internalization linked with membrane binding and stacking due to the hydrophobicity of the triphenylamine part. We previously showed the importance of hydrophobicity in the case of our dimers of conjugated porphyrins.[39]

CONCLUSION

During this study, five potential 2PA photosensitizers were synthesized. Their scaffold relies on the electronic conjugation of a porphyrin core and a triphenylamine part via an alkyne bond. Variations of the *meso* groups (Ph, Ph-TEGOMe and Ph-DEG-mannose) or triphenylamine moieties (Pyr, Bzim) as well as zinc internalization allowed discussion over the spectroscopic properties of the final compounds.

The hybrids displayed high singlet oxygen production quantum yields as well as good 2PA cross sections which encouraged us to test them in 1PA phototoxicity assays. Unfortunately, the compounds optimized for biological media (**4**, **5**) proved to be inactive against usual cancer line HT-29 as well as retinoblastoma cells Y79. These observations were attributed to poor internalization and stacking due to the hydrophobicity of the compounds.

Further optimization has been undertaken so as to remedy to this problem with the functionalization of the TP part with hydrophilic groups.

EXPERIMENTAL SECTION

Synthesis. All solvents used were reagent grade. The following reagents have been abbreviated: dimethylformamide (DMF), dimethyl sulfoxide (DMSO), fetal calf serum (FCS), 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT), phosphate buffer saline (PBS), and Dulbecco's modified Eagle's medium (DMEM). 2-[2-(2-Methoxyethoy)ethoxy]ethanol tosylate **14** (CAS [77544-60-6]) was used without purification. Dry MeOH was kept over 3 Å sieves, and dichloromethane was distilled from calcium hydride and kept over 4 Å sieves. DMF was distilled under slow argon flow and kept over 4 Å sieves. Column chromatography was performed with the indicated solvents using silica gel 60 (particle size 0.035−0.070 mm). Precoated plates (SIL G-200, 2 mm) were used for preparative thin-layer chromatography. Yields refer to chromatographically and spectroscopically pure compounds. ^{1}H and ^{13}C NMR spectra were recorded on a spectrometer (300 MHz) at ambient temperature using an internal deuterium lock. Chemical shift values are given in ppm relative to tetramethylsilane (TMS). Acidic impurities in $CDCl_3$ were removed by treatment with anhydrous K_2CO_3. Melting points were performed on a system Kofler.

General Procedures. *Procedure A (Horner−Wadsworth−Emmons Reaction).* Sodium hydride (60% dispersion in mineral oil) was washed with cyclohexane and introduced in a previously dried flask purged with argon. The phosphonate was added and the mixture stirred for 10−15 min at room temperature. The aldehyde, dissolved in dry THF, was then added to the phosphonate via syringe. After completion, the mixture was partitioned between H_2O and CH_2Cl_2. The aqueous phase was extracted three times with CH_2Cl_2. The organic phases were reunited, washed with water, and dried with anhydrous magnesium sulfate. After filtration, the solvents were evaporated, and the crude product was purified by chromatography to afford the desired products.

Procedure B1 (Sonogashira Coupling with TMSA). The bromozinc derivative was combined with *trans*-dichlorobis(triphenylphosphine-palladium(II) and copper(I) iodide. The flask was purged with argon for 10 min. The solids were dissolved in dry THF and dry triethylamine. The solution was deoxygenated with argon for 5 min and then frozen at 77 K (liquid nitrogen). Trimethylsilylacetylene (TMSA) was added, and the frozen mixture was left under argon flow for 1 h. The gas was then allowed to escape, and the crude solution warmed to room temperature was stirred for 16 h under argon after which the reaction was quenched with water. It was then extracted with CH_2Cl_2 and washed three times with water and once with brine. The organic phase was dried over anhydrous magnesium sulfate and filtered, and the solvents were removed under reduced pressure. The crude product purified on silica gel column.

Procedure B2 (Trimethylsilyl Deprotection Followed by Sonogashira Coupling). To a solution of the silylated derivative in dry THF and/or dry CH_2Cl_2 was slowly added a solution of tetrabutylammonium fluoride (TBAF) 1 M solution in THF. A spatula of $CaCl_2$ was added to quench the reaction after 30 min at 20 °C. The organic layer was concentrated under vacuum, the residue was dissolved in CH_2Cl_2, and the solution was washed with water, dried over anhydrous magnesium sulfate, filtered, and then concentrated under vacuum. The generated product was combined with copper(I) iodide and dichlorobis(triphenylphosphine)palladium(II) in anhydrous THF and dry Et_3N. The reaction mixture was stirred at room temperature overnight. The solution diluted in CH_2Cl_2 was washed with water, the aqueous layer was extracted with CH_2Cl_2, and the combined organic layers were washed with water, dried over anhydrous magnesium sulfate, filtered, and concentrated under vacuum. The products were purified by silica gel chromatography.

Procedure C (Heck Coupling). The trimethylsilyl-protected compound was dissolved in CH_2Cl_2, and TBAF (1 M in THF) was added. The solution was stirred at room temperature for 15 min followed by addition of anhydrous $CaCl_2$. The mixture was stirred for 10 min and filtered, and the solvents were evaporated. The crude product was combined with the adequate iodo or bromo derivative, tris(dibenzylideneacetone)dipalladium(0), and triphenylarsine in anhydrous THF and dry Et_3N under argon. Depending on the reactants, the reaction mixture was allowed to sit at room temperature or heated to reflux under argon. The solvents were evaporated under reduced pressure after which the crude product was taken up in CH_2Cl_2, washed with water, dried over anhydrous magnesium sulfate, and filtered. The solvents were removed under vacuum, and the crude product was purified by chromatography and/or crystallization.

Procedure D (Deprotection of the Alkoxy Groups). The alkoxy-protected porphyrin was dissolved in dry CH_2Cl_2 under argon and cooled to −30 °C upon which boron tribromide BBr_3 was added cautiously. The temperature was maintained between −30 and −20 °C for 20 min. The mixture was slowly poured into a water−ice bath after which a solution of aqueous ammonia was added until the color

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change from green to purple was complete. The mixture was extracted with AcOEt, washed three times with water, dried over anhydrous magnesium sulfate, and filtered. Solvents were removed under reduced pressure and the solid residues dried under vacuum. Crystallization in acetone/*n*-heptane allowed the isolation of the deprotected compound.

Procedure E (Bromination of the Meso Position of Porphyrins). The porphyrin was dissolved in $CHCl_3$ and pyridine, and the resulting solution was cooled to 0 °C. *N*-Bromosuccinimide (NBS) was added and the mixture stirred for 15 min while slowly being warmed to room temperature. Acetone was then introduced to quench the remaining NBS. The solvents were removed under reduced pressure. The remaining solid was then washed with water and dried.

Procedure F (Metalation). The porphyrin was dissolved in $CHCl_3$ or acetone. A solution of zinc(II) acetate in methanol was added, and the mixture was heated at reflux for 5 min. After the mixture was cooled to room temperature and solvents were removed under reduced pressure, the solid residue was taken up in CH_2Cl_2 and washed with water. The organic phase was dried over anhydrous magnesium sulfate and filtered, and the solvents were removed under reduced pressure. The expected compound was then dried under vacuum.

Procedure G (Williamson's Reaction). A mixture of hydroxylated porphyrins, bromo-polyethylene glycol derivatives, and cesium carbonate in dry DMF was stirred under argon at room temperature during 24 h. The mixture was concentrated under vacuum and taken up with a mixture of water and ethyl acetate (2/1, v/v), and the organic layer was separated. The aqueous layer was extracted with ethyl acetate. The combined organic extracts were washed with water (2×), dried over magnesium sulfate, and filtered, and the solvent was evaporated under vacuum. The product was purified by three crystallizations from CH_2Cl_2/*n*-heptane and then purified by preparative chromatography.

*4-Bromo-N,N-bis(4-((E)-2-(pyridin-4-yl)vinyl)phenyl)aniline (TP_{Br}-2Py, **8a**).* Compound **8a** was obtained following procedure A with diethyl[(pyridin-4-yl)methyl]phosphonate (900 mg, 3.9 mmol, 3 equiv) in dry THF (15 mL), NaH (110 mg, 4.6 mmol, 3.5 equiv), and **TP_{Br}-2CHO** (500 mg, 1.32 mmol, 1 equiv) in dry THF (15 mL). The residue was purified over a silica gel column eluted with CH_2Cl_2/*i*-PrOH (95/5, v/v): 322 mg of yellow solid was isolated (θ_f = 176 °C, 46% yield); ^{1}H NMR ($CDCl_3$, 300 MHz) 8.55 (d, *J* = 5.1 Hz, 4H), 7.44 (d, *J* = 8.6 Hz, 4H), 7.40 (d, *J* = 8.8 Hz, 2H), 7.34 (d, *J* = 5.8 Hz, 4H), 7.26 (d, *J* = 15.8 Hz, 2H), 7.09 (d, *J* = 8.6 Hz, 4H), 7.02 (d, *J* = 8.8 Hz, 2H), 6.92 (d, *J* = 16.3 Hz, 2H); ^{13}C NMR ($CDCl_3$, 75.3 MHz) 150.1, 147.3, 146.0, 144.8, 132.5, 132.3, 131.2, 128.2, 126.4, 124.7, 124.0, 120.7, 116.4; MS (ESI$^+$) *m*/*z* $[M + H]^+$ calcd for $C_{32}H_{25}BrN_3$ 530.1, found 530.1; HRMS (MALDI-TOF) (*m*/*z*) $[M]^+$ calcd for $C_{32}H_{24}BrN_3$ 529.1154, found 529.1148.

*4-Iodo-N,N-bis(4-((E)-2-(pyridin-4-yl)vinyl)phenyl)aniline (TP_I-2Py, **8b**).* Following procedure A, the title product was obtained from **TP_I-2CHO** (500 mg, 1.32 mmol, 1 equiv), diethyl[(pyridin-4-yl)methyl]phosphonate (907 mg, 3.96 mmol, 3 equiv), and NaH (110 mg, 4.62 mmol, 3.5 equiv) in dry THF (30 mL). The crude product was purified by silica gel chromatography eluted by a mixture of CH_2Cl_2/*i*-PrOH (95/5, v/v) and yielded 488 mg of the desired compound as an orange powder (θ_f = 117 °C, 64% yield): ^{1}H NMR ($CDCl_3$, 300 MHz) 8.56 (s, 4H), 7.58 (d, *J* = 8.7 Hz, 2H), 7.44 (d, *J* = 8.5 Hz, 4H), 7.35 (d, *J* = 4.7 Hz, 4H), 7.26 (d, *J* = 16.3 Hz, 2H), 7.09 (d, *J* = 8.5 Hz, 4H), 6.95−6.88 (m, 4H); ^{13}C NMR ($CDCl_3$, 75.3 MHz) 150.2, 147.2, 146.7, 144.7, 138.5, 132.3, 131.3, 128.2, 126.6, 124.8, 124.1, 120.7, 86.8; MS (ESI$^+$) *m*/*z* $[M + H]^+$ calcd for $C_{32}H_{25}IN_3$ 578.1, found 578.0.

*4,4′-(4-((Trimethylsilyl)ethynyl)phenylazanediyl)dibenzaldehyde (TP_{YSi}-2CHO, **9**).* Following procedure B1, TP_I-2CHO (630 mg, 1.47 mmol, 1 equiv) was combined with $Pd_2(PPh_3)_2Cl_2$ (105 mg, 0.15 mmol, 0.1 equiv) and CuI (28 mg, 0.15 mmol, 0.1 equiv) in dry THF (10 mL). TMSA (1.25 mL, 8.8 mmol, 6.0 equiv) and dry Et_3N (10 mL) were added via syringe. The reaction was purified by flash chromatography over silica (eluted with ethyl acetate/cyclohexane 30:70, v/v). The title product (533 mg) was obtained as a yellow powder (θ_f = 120 °C, 91% yield): ^{1}H NMR ($CDCl_3$, 300 MHz) 9.90 (s, 2H), 7.79 (d, *J* = 8.5 Hz, 4H), 7.46 (d, *J* = 8.4 Hz, 2H), 7.19 (d, *J* = 8.5 Hz, 4H), 7.09 (d, *J* = 8.4 Hz, 2H), 0.25 (s, 9H); ^{13}C NMR ($CDCl_3$, 75.3 MHz) 190.5, 151.6, 145.6, 133.7, 131.8, 131.4, 126.1, 123.4, 120.5, 104.2, 95.3, 0.0; MS (ESI$^+$) *m*/*z* $[M + Na]^+$ calcd for $C_{25}H_{23}NNaO_2Si$ 420.0, found 420.0; HRMS (MALDI-TOF) (*m*/*z*) $[M]^+$ calcd for $C_{25}H_{23}NO_2Si$ 397.1498, found 397.1506.

*4,4′-(4-Ethynylphenylazanediyl)dibenzaldehyde (TP_Y-2CHO, **10**).* Compound **9** (208 mg, 0.52 mmol) in CH_2Cl_2 (40 mL) was deprotected at 0 °C with 0.62 mL of TBAF (1 M in THF). The reaction was stirred at 0 °C for 30 min. The solution was filtered over a short pad of silica and eluted with a mixture CH_2Cl_2/*i*-PrOH (9/1, v/v). The solvents removed under vacuum to afford 200 mg (quant) of the title compound as a dark yellow solid (θ_f = 99 °C): ^{1}H NMR ($CDCl_3$, 300 MHz) 9.84 (s, 2H), 7.72 (d, *J* = 8.4 Hz, 4H), 7.41 (d, *J* = 8.3 Hz, 2H), 7.12 (d, *J* = 8.4 Hz, 4H), 7.04 (d, *J* = 8.3 Hz, 2H), 3.05 (s, 1H); ^{13}C NMR ($CDCl_3$, 75.3 MHz) 188.9, 150.0, 144.4, 132.3, 130.2, 129.8, 124.5, 121.8, 117.8, 81.3, 76.5; MS (ESI$^+$) *m*/*z* $[M + H]^+$ calcd for $C_{22}H_{16}NO_2$ 326.1, found 326.2; HRMS (MALDI-TOF) (*m*/*z*) $[M]^+$ calcd for $C_{22}H_{15}NO_2$ 325.1103, found 325.1091.

*PTP_M-2CHO (**12**).* According to procedure C, a flask was charged with **TP_Y-2CHO** (201 mg, 0.47 mmol, 2 equiv), **TPP_MBr** (150 mg, 0.24 mmol, 1 equiv), $Pd_2(dba)_3$ (17 mg, 0.07 mmol, 0,3 equiv), and $AsPh_3$ (110 mg, 0.36 mmol, 1.5 equiv), and purged for 10 min with argon. The solids were dissolved in dry THF (20 mL), and the solution was deoxygenated for 20 min before the addition of dry Et_3N (4 mL). The reaction was stirred for 20 h at room temperature. The solvents were removed under reduced pressure, and the residue was purified by silica gel column chromatography ($CHCl_3/Et_2O$, 98/2, v/v) to afford 250 mg (84%) of **PTP_M-2CHO** as a dark blue-green powder: UV−vis in CH_2Cl_2 λ_{max} nm (ε mM^{-1} cm^{-1}) 444 (419.0), 569 (26.7), 618 (43.5); ^{1}H NMR ($CDCl_3$, 300 MHz) 9.93 (s, 2H), 9.83 (d, *J* = 4.6 Hz, 2H), 8.95 (d, *J* = 4.6 Hz, 2H), 8.80 (s, 4H), 8.24 (m, 6H), 8.10 (d, J = 8.5 Hz, 2H), 7.87 (d, *J* = 8.6 Hz, 4H), 7.79 (m, 9H), 7.40 (d, J = 8.5 Hz, 2H), 7.35 (d, *J* = 8.5 Hz, 4H); ^{13}C NMR ($CDCl_3$, 75.3 MHz) 187.5, 150.3, 149.7, 148.6−147.8, 144.0, 141.4−141.3, 132.5−132.4, 131.1, 130.5, 130.4, 129.7−129.4, 129.0, 128.3, 125.5, 124.5−124.4, 121.5, 120.7−119.7, 97.0, 93.1, 92.0; MS (MALDI-TOF) *m*/*z* $[M]^+$ calcd for $C_{60}H_{37}N_5O_2Zn$ 923.22, found 923.21; HRMS (MALDI-TOF) (*m*/*z*) $[M]^+$ calcd for $C_{60}H_{37}N_5O_2Zn$ 923.2239, found 923.2273.

*PTP-2CHO (**13**).* Compound **12** (250 mg, 0.27 mmol) was dissolved in $CHCl_3$ (20 mL) and introduced in a separatory funnel. TFA (1 mL) was added, and the funnel was shaken for a few minutes. The mixture was washed with water until pH 7 was reached. The organic phase was dried over magnesium sulfate and filtered, and the solvents were removed under reduced pressure. Purification was achieved by preparative TLC (AcOEt/cyclohexane, 30/70, v/v) to afford the desired compound as a blue solid (147 mg, 63%): UV−vis in CH_2Cl_2 λ_{max} nm (ε mM^{-1} cm^{-1}): 437 (385.3), 532 (24.9), 581 (49.5), 615 (15.8), 670 (23.9); ^{1}H NMR ($CDCl_3$, 300 MHz) 9.95 (s, 2H), 9.72 (d, *J* = 4.8 Hz, 2H), 8.92 (d, *J* = 4.7 Hz, 2H), 8.76 (s, 4H), 8.19 (m, 6H), 8.02 (d, *J* = 8.6 Hz, 2H), 7.86 (d, *J* = 8.6 Hz, 4H), 7.78 (m, *J* = 6.2 Hz, 9H), 7.33 (m, *J* = 8.4 Hz, 6H), −2.31 (s, 2H); ^{13}C NMR ($CDCl_3$, 75.3 MHz) 190.7, 141.7, 134.5, 133.3, 131.5, 127.9, 126.8, 126.5, 123.5, 121.2; MS (MALDI-TOF) *m*/*z* $[M]^+$ calcd for $C_{60}H_{39}N_5O_2$ 861.31, found 861.36.

*5-H-10,15,20-Tri-p-phenol Porphyrin ($TPP(OH)_3$, **15**).* Following procedure D, **$TPP(OiPr_2OH)_3$**[44] (800 mg, 1.19 mmol, 1 equiv) was dissolved in dry CH_2Cl_2 (100 mL), then BBr_3 (2.26 mL, 23.8 mmol, 20 equiv) was added. **$TPP(OH)_3$** was precipitated from AcOEt with *n*-heptane to afford 595 mg of purple powder (yield 85%): UV−vis in acetone λ_{max} nm (ε mM^{-1} cm^{-1}) 412 (420), 509 (15.3), 545 (7.8), 586 (4.5), 642 (3.3); ^{1}H NMR (acetone-d_6, 300 MHz) 10.17 (s, 1H), 9.30 (d, *J* = 4.6 Hz, 2H), 8.98 (d, *J* = 4.6 Hz, 2H), 8.3 (m, 4H), 8.03 (t, *J* = 8.6 Hz, 6H), 7.28 (d, *J* = 8.4 Hz, 6H), −2.98 (s, 2H); ^{13}C NMR (acetone-d_6, 75.3 MHz) 170.2, 170.1, 158.7, 135.7−135.2, 134.4, 131.45, 120.3, 119.3, 113.1, 112.7, 97.7 (C-20); MS (MALDI-TOF) *m*/*z* $[M]^+$ calcd for $C_{38}H_{26}N_4O_3$ 586.20, found 586.21; HRMS

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(MALDI-TOF) (m/z) $[M]^+$ calcd for $C_{38}H_{26}N_4O_3$ 586.2005, found 586.2005.

*5-Bromo-10,15,20-tri-p-phenol Porphyrin (TPP(OH)$_3$Br, **16**).* The title compound was obtained through procedure E from **TPP(OH)$_3$** (183 mg, 0.23 mmol, 1 equiv). **TPP(OH)$_3$Br** was isolated as a dark blue powder (149 mg, 97%) and used without purification: UV−vis in CH_2Cl_2 λ_{max} nm (ε mM^{-1} cm^{-1}) 424 (218.7), 523 (9.6), 560 (7.1), 600 (3.2), 658 (3.8); ^1H NMR (acetone-d_6, 300 MHz) 9.60 (d, J = 4.8 Hz, 2H), 8.94 (d, J = 4.5 Hz, 2H), 8.89 (s, 4H), 8.03 (d, J = 8.4 Hz, 6H), 7.33−7.28 (m, 6H), 2.96 (broad s, 3H), −2.74 (s, 2H); ^{13}C NMR (acetone-d_6, 75.3 MHz) 158.6, 136.6, 136.4, 133.7, 133.5, 122.4, 121.9, 114.8, 102.2; MS (MALDI-TOF) m/z $[M]^+$ calcd for $C_{38}H_{25}BrN_4O_3$ 664.11, found 664.11; HRMS (MALDI-TOF) (m/z) $[M]^+$ calcd for $C_{38}H_{25}BrN_4O_3$ 664.1110, found 664.1109.

*5-Bromo-10,15,20-tri-p-phenol Porphyrin Zinc Complex (TPP$_M$(OH)$_3$Br, **17**).* Procedure F afforded the title compound starting from **TPP(OH)$_3$Br** (130 mg, 195 mmol, 1 equiv) and zinc(II) acetate (179 mg, 977 mmol, 5.0 equiv) in 70 mL of acetone and 30 mL of MeOH. The solvents were evaporated, and **17** was isolated as a purple powder (120 mg, 84% yield): UV−vis in CH_2Cl_2 λ_{max} nm (ε mM^{-1} cm^{-1}) 430 (111.8), 566 (4.4), 640 (7.2); ^1H NMR (acetone-d_6, 300 MHz) 9.70 (d, J = 4.7 Hz, 2H), 9.00 (d, J = 4.7 Hz, 2H), 8.91 (s, 4H), 8.05−8.00 (m, 6H), 7.29 (dd, J_1 = 11.4 Hz, J_2 = 4.9 Hz, 6H), 2.77 (s, 3H); ^{13}C NMR (acetone-d_6, 75.3 MHz) 158.1, 151.8−150.1, 136.4, 136.3, 135.0, 134.9, 133.8, 132.9, 132.8, 122.7, 122.3, 114.4, 103.6, 100.9; MS (MALDI-TOF) m/z $[M]^+$ calcd for $C_{38}H_{23}BrN_4O_3Zn$ 726.02, found 726.02; HRMS (MALDI-TOF) (m/z) $[M]^+$ calcd for $C_{38}H_{23}BrN_4O_3Zn$ 726.0245, found 726.0274.

*5-Bromo-10,15,20-tri(p-2-(2-(2-methoxyethoy)ethoxy)-ethanoxyphenylporphyrin Zinc Complex (TPP$_M$(OTEG)$_3$Br, **18**).* According to procedure G, **TPP$_M$(OH)$_3$Br** (660 mg, 0.87 mmol, 1 equiv), TEGOTs (4.15 g, 13.05 mmol, 15 equiv), and Cs_2CO_3 (12.76 g, 40 mmol, 45 equiv) reacted in anhydrous DMF (100 mL) to afford **18** as a purple solid after recrystallization from CH_2Cl_2/n-heptane (913 mg, 88%): UV−vis in CH_2Cl_2 λ_{max} nm (ε mM^{-1} cm^{-1}) 429 (207.0), 564 (7.5), 605 (5.0); ^1H NMR ($CDCl_3$, 300 MHz) 9.74 (d, J = 4.6 Hz, 2H), 8.98 (d, J = 4.6 Hz, 2H), 8.88 (s, 4H), 8.07−8.04 (m, 6H), 7.20−7.17 (m, 6H), 4.19 (m, 6H), 3.81 (m, 6H), 3.62−3.61 (m, 6H), 3.51−3.44 (m, 12H), 3.35 (m, 6H), 1.43 (s, 9H); ^{13}C NMR ($CDCl_3$, 75.3 MHz) 158.4, 150.9, 149.6, 135.4, 132.2, 121.3, 112.7, 71.7−67.5, 58.9; MS (MALDI-TOF) m/z $[M]^+$ calcd for $C_{59}H_{65}BrN_4O_{12}Zn$ 1164.31, found 1164.31; HRMS (MALDI-TOF) (m/z) $[M]^+$ calcd for $C_{59}H_{65}BrN_4O_{12}Zn$ 1164.3074, found 1164.3014.

*5-Allyl-10,15,20-tri(p-2-(2-(2-methoxyethoy)ethoxy)-ethanoxyphenylporphyrin Zinc Complex. TPP$_M$(OTEG)$_3$YSi (**19**).* Following procedure B1, **TPP$_M$(OTEG)$_3$Br** (0.80 g, 0.69 mmol, 1 equiv) was combined with TMSA (390 mg, 4.1 mmol, 6 equiv), $Pd(PPh_3)_2Cl_2$ (47 mg, 69 mmol, 0.1 equiv), and CuI (12 mg, 69 mmol, 0.1 equiv) in dry THF (15 mL) and dry Et_3N (2.5 mL). The title compound was obtained after purification over silica gel chromatography (CH_2Cl_2/MeOH, 99/1, v/v) as a blue powder (560 mg, 69% yield): UV−vis in CH_2Cl_2 λ_{max} nm (ε mM^{-1} cm^{-1}) 435 (259.8), 572 (9.2), 619 (11.6); ^1H NMR ($CDCl_3$, 300 MHz) 9.72 (d, J = 4.6 Hz, 2H), 8.96 (d, J = 4.6 Hz, 2H), 8.84 (m, 4H), 8.03 (m, 6H), 7.09 (m, 6H), 4.01 (m, 6H), 3.62 (m, 6H), 3.42 (m, 6H), 3.31 (m, 6H), 3.23 (m, 6H), 3.14 (m, 6H), 2.99 (s, 9H), 0.62 (s, 9H); ^{13}C NMR ($CDCl_3$, 75.3 MHz) 157.8, 152.0, 150.3, 149.6, 134.9, 135.2, 132.3, 131.6, 130.2, 121.0, 112.2, 98.5, 95.6, 93.7, 71.0−69.0, 58.2, 0.0; MS (MALDI-TOF) m/z $[M]^+$ calcd for $C_{64}H_{74}N_4O_{12}SiZn$ 1182.44, found 1182.44.

*PTP$_M$(OTEG)$_3$-2CHO (**20**).* Following procedure C, **19** (550 mg, 0.46 mmol, 1 equiv) was deprotected with TBAF (506 μL at 1 M in THF, 0.56 mmol, 1.2 equiv) in THF (40 mL) and CH_2Cl_2 (8 mL) and was then reacted with **TP$_I$-2CHO** (370 mg, 0.87 mmol, 1.9 equiv), Pd_2dba_3 (30 mg, 0.13 mmol, 0.3 equiv), and $AsPh_3$ (200 mg, 0.65 mmol, 1.5 equiv) in dry THF (20 mL) and dry Et_3N (20 mL). Silica gel chromatography eluted with CH_2Cl_2/EtOH (99/1 to 95/5, v/v) afforded 650 mg of **20** (99% yield) as a dark blue powder: UV−vis in CH_2Cl_2 λ_{max} nm (ε mM^{-1} cm^{-1}) 377 (20.0), 450 (203.4), 580 (8.2), 634 (22.3); ^1H NMR ($CDCl_3$, 300 MHz) 9.77 (s, 4H), 8.99 (d, J = 4.6 Hz, 4H), 8.86 (s, 4H), 8.07−8.01 (m, 6H), 7.95 (d, J = 8.5 Hz, 2H), 7.66 (d, J = 8.7 Hz, 4H), 7.14 (m, 6H), 7.05 (d, J = 8.7 Hz, 4H), 4.08 (m, 6H), 3.70 (m, 6H), 3.51 (m, 6H), 3.42−3.31 (m, 12H), 3.23 (m, 6H), 3.09 (s, 9H); ^{13}C NMR ($CDCl_3$, 75.3 MHz) 190.5, 158.3, 152.1, 151.5, 150.9, 150.3, 150.0, 135.4, 135.2, 133.1, 131.5, 131.3, 126.5, 123.1, 121.7, 112.7, 99.0, 95.1, 94.2, 71.6−67.4, 58.8; MS (MALDI-TOF) m/z $[M]^+$ calcd for $C_{81}H_{79}N_5O_{14}Zn$ 1409.49, found 1409.50; HRMS (MALDI-TOF) (m/z) $[M]^+$ calcd for $C_{81}H_{79}N_5O_{14}Zn$ 1409.4915, found 1409.4908.

*5,10,15-Tri[(p-2′,3′,4′,5′,6′-tetracacetyl-α-D-mannosyloxy-2-ethoxy)-2-ethyloxyphenyl]porphyrin (TPP(DEGManOAc)$_3$, **21**).*[39] *5-Allyl-10,15,20-tri[(p-2′,3′,4′,5′,6′-tetracetyl-α-D-mannosyloxy-2-ethoxy)-2-ethyloxyphenyl]porphyrin Zinc Complex (TPP$_M$(DEGManOAc)$_3$YSi, **22**).*[39] *5-Allyl-10,15,20-tri(p-hydroxyphenyl)porphyrin Zinc Complex (TPP$_M$(OH)$_3$YSi, **23**).* Following procedures E, F, and B1, starting from **15**, the expected compound **23** was isolated after purification by preparative TLC eluted with CH_2Cl_2/EtOH (95/5, v/v) as a dark blue solid (32 mg, 59%): UV−vis in CH_2Cl_2 λ_{max} nm (ε mM^{-1} cm^{-1}) 431 (122.3), 562 (5.7), 609 (4.4); ^1H NMR (acetone-d_6, 300 MHz) 9.62 (d, J = 4.6 Hz, 2H), 8.90 (d, J = 4.6 Hz, 2H), 8.81−8.77 (m, 4H), 7.92 (m, 6H), 7.19 (m, 8.3 Hz, 6H), 0.60 (s, 9H); ^{13}C NMR (acetone-d_6, 75.3 MHz) 158.1, 153.1−151.0, 136.3, 136.2−132.2, 122.6, 114.4, 55.4, 0.5; MS (MALDI-TOF) m/z $[M]^+$ calcd for $C_{43}H_{32}N_4O_3SiZn$ 744.15, found 744.11; HRMS (MALDI-TOF) (m/z) $[M]^+$ calcd for $C_{43}H_{32}N_4O_3SiZn$ 744.1535, found 744.1547.

*PTP$_M$(OH)$_3$-2CHO (**24**).* The title compound was obtained following procedure C starting from **23** (265 mg, 0.36 mmol, 1 equiv) and TBAF (0.47 mL at 1 M in THF, 0.47 mmol, 1.3 equiv) in dry CH_2Cl_2 (5 mL) and dry THF (25 mL) first followed by **7b** (303 mg, 0.71 mmol, 2 equiv) in the presence of Pd_2dba_3 (25 mg, 0.11 mmol, 0.3 equiv) and $AsPh_3$ (165 mg, 0.54 mmol, 1.5 equiv) in dry THF (20 mL) and dry Et_3N (10 mL). Purification over silica gel eluted with CH_2Cl_2/EtOH (95/5, v/v) afforded the desired compound with a 69% yield (243 mg) as a dark blue powder: UV−vis in THF λ_{max} nm (ε mM^{-1} cm^{-1}) 446 (173.7), 564 (7.3), 630 (17.3); ^1H NMR (acetone-d_6, 300 MHz) 9.69 (s, 2H), 9.65 (d, J = 4.6 Hz, 2H), 8.84 (d, J = 4.6 Hz, 2H), 8.70 (s, 4H), 7.90 (d, J = 8.4 Hz, 2H), 7.84 (t, J = 7.9 Hz, 6H), 7.60 (d, J = 8.5 Hz, 4H), 7.10 (m, 11H), 7.01 (d, J = 8.5 Hz, 4H); ^{13}C NMR (acetone-d_6, 75.3 MHz) 191.2, 158.1, 152.9, 152.5, 152.4, 151.8, 151.1, 150.9, 146.5, 138.2, 136.4, 134.9, 132.9, 132.4, 132.0, 129.1, 127.5, 126.1, 124.2, 114.5, 99.3, 95.9, 94.8; MS (MALDI-TOF) m/z $[M]^+$ calcd for $C_{60}H_{37}N_5O_5Zn$ 971.21, found 971.20; HRMS (MALDI-TOF) (m/z) $[M]^+$ calcd for $C_{60}H_{37}N_5O_5Zn$ 971.2086, found 971.2088.

*PTP$_M$(DEG(Man(OAc)$_4$)$_3$-2CHO (**25**).* The title compound was obtained through procedure G from **24** (55 mg, 0.06 mmol, 1 equiv) with Cs_2CO_3 (159 mg, 2.70 mmol, 45 equiv) and [(2-bromoethoxy)-*O*-ethoxy]-*O*-2′,3′,4′,6′-tetraacetyl-α-D-mannose (427 mg, 0.90 mmol, 15 equiv). Compound **25** was isolated after purification over silica gel eluted with cyclohexane/AcOEt (7/3, v/v) as a dark blue powder (19 mg, 70%): UV−vis in CH_2Cl_2 λ_{max} nm (ε mM^{-1} cm^{-1}) 447 (136.5), 576 (6.8), 628 (17.4); ^1H NMR ($CDCl_3$, 300 MHz) 9.80 (s, 2H), 9.01 (s, 4H), 8.87 (s, 4H), 8.08 (s, 6H), 7.97 (d, J = 7.9 Hz, 2H), 7.73 (d, J = 7.7 Hz, 2H), 7.30 (m, 8H), 7.10 (d, J = 7.4 Hz, 2H), 5.42−5.33 (m, 9H), 4.96 (s, 3H), 4.40−4.31 (m, 9H), 4.17 (s, 6H), 4.02 (s, 6H), 3.85 (s, 12H), 2.16 (s, 9H), 2.11 (s, 9H), 2.03 (s, 9H), 1.99 (s, 9H); ^{13}C NMR ($CDCl_3$, 75.3 MHz) 170.7, 170.0, 158.5, 152.8, 150.9, 135.5, 133.0−129.7, 112.8, 97.8, 70.4, 70.0, 69.6, 69.2, 68.5, 67.5, 66.2, 62.5, 20.9, 20.7; MS (MALDI-TOF) m/z $[M + H]^+$ calcd for $C_{114}H_{116}N_5O_{38}Zn$ 2227.52, found 2227.65; HRMS (MALDI-TOF) (m/z) $[M]^+$ calcd for $C_{114}H_{115}N_5O_{38}Zn$ 2226.6545, found 2226.6681.

*5-Trimethylsilyl-10,15,20-tri(p-isopropyloxyphenyl)porphyrin Zinc Complex (TPP$_M$(OiPr)$_3$YSi, **26**).* Following procedure B1, **TPP$_M$(OiPr)$_3$Br** (691 mg, 0.81 mmol, 1 equiv) reacted with TMSA (685 mL, 4.85 mmol, 6 equiv) in the presence of $Pd(PPh_3)_2Cl_2$ (57 mg, 0.08 mmol, 0.1 equiv) and CuI (15 mg, 0.08 mmol, 0.1 equiv) in dry THF (20 mL) and dry Et_3N (1 mL). The desired compound was isolated as a dark blue solid after purification over silica gel column

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eluted with CH_2Cl_2 (576 mg, 82%): UV−vis in CH_2Cl_2 λ_{max} nm (ε mM^{-1} cm^{-1}): 433 (436.9), 567 (16.3), 612 (14.4); ^1H NMR ($CDCl_3$, 300 MHz) 9.71 (d, J = 4.4 Hz, 2H), 9.00 (d, J = 4.4 Hz, 2H), 8.88 (s, 4H), 8.06 (d, J = 8.0 Hz, 6H), 7.25 (m, 6H), 4.83 (sept, J = 5.7 Hz, 3H), 1.54 (d, J = 5.5 Hz, 18H), 0.61 (s, 9H); ^{13}C NMR ($CDCl_3$, 75.3 MHz) 158.0, 153.0−150.6, 136.0, 135.9, 135.4−135.3, 133.4−132.1, 131.2, 123.3, 122.2, 114.3−114.2, 108.2, 101.5, 70.6, 22.8, 0.9; MS (MALDI-TOF) m/z $[M]^+$ calcd for $C_{52}H_{50}N_4O_3SiZn$ 870.29, found 870.26; HRMS (MALDI-TOF) (m/z) $[M]^+$ calcd for $C_{52}H_{50}N_4O_3SiZn$ 870.2944, found 870.2916.

*$PTP_M(OiPr)_3$-2CHO (**27**).* Following procedure H, **26** (370 mg, 0.42 mmol, 1 equiv) was put to reaction first with TBAF (1 M in THF) in dry CH_2Cl_2 (7 mL) and dry THF (36 mL). The crude product was combined with **TP_{Br}-2CHO** (177 mg, 0.47 mmol, 1.1 equiv), Pd_2dba_3 (30 mg, 0.13 mmol, 0.3 equiv), and $AsPh_3$ (193 mg, 0.63 mmol, 1.5 equiv) in dry THF (53 mL) and dry Et_3N (11 mL). The crude was purified twice over a silica gel column eluted with CH_2Cl_2/EtOH (100/1 to 98/2, v/v) to afford the desired compound as a dark blue solid (88 mg, 19%): UV−vis in CH_2Cl_2 λ_{max} nm (ε mM^{-1} cm^{-1}) 446 (231.1), 522 (3.2), 572 (12.1), 624 (25.9); ^1H NMR ($CDCl_3$, 300 MHz) 9.66 (d, J = 4.6 Hz, 2H), 9.56 (s, 2H), 8.98 (d, J = 4.6 Hz, 2H), 8.91 (d, J = 0.9 Hz, 4H), 8.04 (d, J = 8.4 Hz, 6H), 7.92 (d, J = 8.4 Hz, 2H), 7.60 (d, J = 8.6 Hz, 4H), 7.24 (d, J = 10.2 Hz, 6H), 7.15 (d, J = 8.5 Hz, 2H), 7.07 (d, J = 8.5 Hz, 4H), 4.91−4.69 (m, 3H,) 1.55 (d, J = 6.0 Hz, 18H); ^{13}C NMR ($CDCl_3$, 75.3 MHz) 190.4, 157.6, 152.0, 151.5, 150.9, 150.3, 150.0, 145.1, 135.6, 134.7, 134.6, 133.2, 132.9, 132.3, 131.9, 131.4, 131.3, 130.3, 126.4, 123.1, 121.9, 121.7, 113.9, 99.1, 95.2, 93.8, 70.1, 22.3; MS (MALDI-TOF) m/z $[M]^+$ calcd for $C_{69}H_{55}N_5O_5Zn$ 1097.35, found 1097.36; HRMS (MALDI-TOF) (m/z) $[M]^+$ calcd for $C_{69}H_{55}N_5O_5Zn$ 1097.3495 found 1097.3524.

*$PTP_M(OiPr)_3$-2Py (**28**).* Compound **27** (86 mg, 0.078 mmol, 1 equiv) was converted to the title compound according to procedure A with $PyrCH_2PO(OEt)_2$ (72 mg, 0.313 mmol, 4.0 equiv) and NaH (8 mg, 0.351 mmol, 4.5 equiv). Purification over a silica gel column eluted with CH_2Cl_2/EtOH (99/1 to 95/5, v/v with a very weak gradient) afforded the desired compound **28** as a dark blue solid (20 mg, 0.016 mmol, 21%): UV−vis in THF λ_{max} nm (ε mM^{-1} cm^{-1}) 430 (110.3), 521 (3.0), 592 (6.4), 681 (2.8); ^1H NMR ($CDCl_3$, 300 MHz) 9.66 (d, J = 4.2 Hz, 2H), 8.91 (d, J = 5.0 Hz, 2H), 8.80 (s, 4H), 8.03 (d, J = 8.4 Hz, 6H), 7.81 (d, J = 9.1 Hz, 2H), 7.19 (m, 10H), 7.14−7.07 (m, 6H), 6.93 (d, J = 9.0 Hz, 4H), 6.73 (d, J = 10.2 Hz, 2H), 6.31 (m, 6H), 4.79 (m, 3H), 1.51 (d, J = 5.9 Hz, 18H); ^{13}C NMR ($CDCl_3$, 75.3 MHz) 157.4, 152.1, 150.8, 150.2, 149.9, 147.3, 146.7, 144.7, 135.6, 135.4, 132.8, 132.7, 130.7, 128.1, 124.7, 124.0, 121.5, 119.9, 113.7, 98.5, 95.3, 94.1, 70.1, 22.3; MS (MALDI-TOF) m/z $[M]^+$ calcd for $C_{81}H_{65}N_7O_3Zn$ 1247.44, found 1247.43.

*$PTP(DEGMan(OAc)_4)_3$-2Py (**30**).* Following procedure D, **28** was reacted with BBr_3 (0.05 mL, 0.48 mmol, 30 equiv) in dry CH_2Cl_2 (2.5 mL) to afford compound **29**, which was directly engaged in procedure G with Cs_2CO_3 (1.07 g, 3.28 mmol, 45 equiv) and [(2-bromoethoxy)-*O*-ethoxy]-*O*-2′,3′,4′,6′-tetraacetyl-α-D-mannose (547 mg, 1.1 mmol, 15 equiv). The reaction was catalyzed with KI (3.6 mg, 0.022 mmol, 0.3 equiv). The expected compound was isolated by recrystallization from CH_2Cl_2/*n*-heptane (35 mg, 20% yield) as a dark blue powder: UV−vis in CH_2Cl_2 λ_{max} nm (ε mM^{-1} cm^{-1}) 450 (210.9), 581 (10.9), 638 (41.3); ^1H NMR ($CDCl_3$, 300 MHz) 9.71 (s, 2H), 8.92 (s, 2H), 8.78 (s, 4H), 8.58 (s, 4H), 8.10 (d, J = 7.7 Hz, 6H), 7.95 (d, J = 7.6 Hz, 2H), 7.53 (d, J = 7.4 Hz, 4H), 7.42−7.14 (m, 15H), 6.98 (d, J = 16.1 Hz, 2H), 5.49−5.28 (m, 9H), 4.99 (s, 3H), 4.41−3.89 (4m, 36H), 2.18 (s, 9H), 2.14 (s, 9H), 2.04 (s, 9H), 2.00 (s, 9H), −2.27 (s, 2H); ^{13}C NMR ($CDCl_3$, 75.3 MHz) 171.1, 170.7, 170.5, 170.4, 170.1, 170.0, 169.8, 158.7, 150.1, 147.2, 147.1, 144.8, 135.6, 134.3, 132.8, 132.4, 131.5, 128.2, 124.8, 124.5, 124.1, 120.7, 118.5, 113.0, 99.5, 97.8, 96.8, 92.2, 70.4, 70.0, 69.6, 69.5, 69.2, 68.5, 68.0, 67.7, 67.5, 67.0, 66.2, 63.6, 62.5, 30.9, 21.0, 20.8, 20.7; MS (MALDI-TOF) m/z $[M]^+$ calcd for $C_{126}H_{127}N_7O_{36}$ 2314.84, found 2314.75.

*$PTP_M(DEGMan(OAc)_4)_3$-2Py (**31**).* Compound **30** (35 mg, 15 μmol, 1 equiv) was metalated following procedure F in the presence of zinc acetate (14 mg, 76 μmol, 5 equiv). Compound **31** was isolated quantitatively as a blue powder and used without purification: UV−vis in DMF λ_{max} nm (ε mM^{-1} cm^{-1}) 450 (79.6), 581 (4.6), 638 (16.1); ^1H NMR ($CDCl_3$, 300 MHz) 9.59 (d, J = 4.3 Hz, 2H), 8.81 (d, J = 3.7 Hz, 2H), 8.71 (s, 4H), 7.98 (d, J = 7.4 Hz, 6H), 7.75 (d, J = 7.9 Hz, 2H), 7.25−7.01 (m, 13H), 6.91 (d, J = 7.5 Hz, 4H), 6.80−6.60 (m, 2H), 6.33 (m, 6H), 5.39−5.20 (m, 4.6 Hz, 9H), 4.90 (s, 3H), 4.28−3.78 (4m, 36H), 2.09 (s, 9H), 2.05 (s, 9H), 1.95 (s, 9H), 1.91 (s, 9H); ^{13}C NMR ($CDCl_3$, 75.3 MHz) 169.7, 169.1, 168.9, 168.8, 157.2, 151.0, 149.7, 149.1, 148.8, 146.3, 145.6, 145.2, 144.0, 134.8, 134.4, 132.0, 131.6, 130.8, 129.6, 127.1, 123.9, 122.9, 122.4, 121.3, 120.3, 118.9, 111.5, 97.5, 96.8, 94.3, 93.0, 69.3, 69.0, 68.8, 68.6, 68.4, 68.1, 67.5, 66.9, 66.6, 66.5, 65.2, 62.6, 61.5, 20.0, 19.9, 19.8, 19.7, 19.7; MS (MALDI-TOF) m/z $[M + H]^+$ calcd for $C_{126}H_{126}N_7O_{36}Zn$ 2377.75, found 2377.70; HRMS (MALDI-TOF) (m/z) $[M]^+$ calcd for $C_{126}H_{125}N_7O_{36}Zn$ 2376.7491, found 2376.7469.

*PTP_M-2Py (**1**).* Compound **1** was synthesized according to procedure F from **12** (50 mg, 54 μmol, 1 equiv), $PyrCH_2PO(OEt)_2$ (37 mg, 162 μmol, 3 equiv), and NaH (8 mg, 189 μmol, 3.5 equiv) in dry THF (2 mL). The crude product was purified by preparative TLC (CH_2Cl_2/MeOH, 95/5, v/v) to afford 35 mg (64%) of the title compound as a dark blue powder: UV−vis in DMSO λ_{max} nm (ε mM^{-1} cm^{-1}) 448 (160.0), 580 (10.6), 634 (30.7); ^1H NMR ($CDCl_3$, 300 MHz) 9.61 (d, J = 4.5 Hz, 2H), 8.79 (d, J = 4.5 Hz, 2H), 8.67 (s, 4H), 8.06 (s, 6H), 7.73 (d, J = 8.3 Hz, 2H), 7.60 (m, 9H), 7.26 (d, J = 8.5 Hz, 2H), 7.08−7.01 (m, 4H), 6.86 (d, J = 8.4 Hz, 4H), 6.81 (d, J = 5.0 Hz, 2H), 6.73 (d, J = 16.3 Hz, 2H), 6.48 (s, 4H), 6.34 (d, J = 16.3 Hz, 2H), 6.09 (m, 2H); ^{13}C NMR ($CDCl_3$, 75.3 MHz) 151.1, 149.4, 148.8, 148.6, 147.3, 146.3, 146.2, 145.3, 143.7, 142.2, 142.1, 135.0, 133.5, 133.4, 131.7, 131.5, 130.9, 130.5, 129.7, 129.3, 127.0, 126.2, 125.3, 123.4, 123.1, 122.7, 122.4, 121.5, 120.6, 119.0, 118.4, 98.0, 94.4, 92.8; MS (MALDI-TOF) m/z $[M]^+$ calcd for $C_{72}H_{47}N_7Zn$ 1073.32, found 1073.31; HRMS (MALDI-TOF) (m/z) $[M]^+$ calcd for $C_{72}H_{47}N_7Zn$ 1073.3184, found 1073.3167.

*PTP-2Py (**2**).* Following procedure F, **13** (60 mg, 70 mmol, 1 equiv) was reacted with $PyrCH_2PO(OEt)_2$ (48 mg, 210 μmol, 3 equiv) and NaH (10 mg, 245 μmol, 3.5 equiv). The crude product was purified over silica gel column chromatography (CH_2Cl_2/EtOH, 95/5, v/v) to yield the title compound as a dark blue solid (30 mg, 42%): UV−vis in DMSO λ_{max} nm (ε mM^{-1} cm^{-1}) 376 (103.5), 435 (210.0), 521 (20.5), 580 (33.2), 669 (15.8); ^1H NMR ($CDCl_3$, 300 MHz) 9.72 (d, J = 4.8 Hz, 2H), 8.90 (d, J = 4.7 Hz, 2H), 8.75 (s, 4H), 8.48 (d, J = 3.7 Hz, 4H), 8.20−8.15 (m, 6H), 7.89 (d, J = 8.6 Hz, 2H), 7.75−7.70 (m, 9H), 7.41 (d, J = 8.5 Hz, 4H), 7.32−7.08 (m, 12H), 6.85 (d, J = 16.3 Hz, 2H), −2.27 (s, 2H); ^{13}C NMR ($CDCl_3$, 75.3 MHz) 150.1, 147.2, 147.1, 144.8, 142.0−141.7, 134.5−134.4, 132.9, 132.4, 131.5, 128.3, 127.9, 126.9, 126.8, 124.8, 124.6, 124.1, 121.8−118.5, 120.7, 99.9, 96.9, 92.2; MS (MALDI-TOF) m/z $[M]^+$ calcd for $C_{72}H_{49}N_7$ 1011.40, found 1011.43; HRMS (MALDI-TOF) (m/z) $[M]^+$ calcd for $C_{72}H_{49}N_7$ 1011.4049, found 1011.4050. Anal. Calcd for $C_{72}H_{49}N_7 \cdot 14H_2O$: C, 68.39; H, 6.14; N, 7.75. Found: C, 68.04; H, 5.98; N, 7.13.

*PTP_M-2Bzim (**3**).* Compound **12** (100 mg, 0.11 mmol, 1 equiv) was dissolved in THF (5 mL) along with (1-methylbenzimidazol-2-ylmethyl)triphenylphosphonium chloride (97 mg, 0.23 mmol, 2.2 equiv) and 1,8-diazabicyclo[5.4.0]undec-7-ene (83 μL, 0.55 mmol, 5 equiv). The mixture was stirred at room temperature in the dark for 15 h. The solvent was removed under reduced pressure and the residue taken up in ethyl acetate, washed with water and then brine, dried over magnesium sulfate, and filtered, and the solvents were removed. The product was purified over silica gel column chromatography (CH_2Cl_2/MeOH, 98/2, v/v) to afford 28 mg (22%) of the desired compound as a dark blue powder: UV−vis in DMSO λ_{max} nm (ε mM^{-1} cm^{-1}) 380 (75.9), 446 (359.3), 584 (30.1), 634 (82.6); ^1H NMR ($CDCl_3$, 300 MHz) 9.62 (d, J = 4.6 Hz, 2H), 8.84 (d, J = 4.6 Hz, 2H), 8.72 (dd, J_1 = 12.3 Hz, J_2 = 4.6 Hz, 4H), 8.15 (d, J = 8.5 Hz, 2H), 8.08 (d, J = 8.4 Hz, 4H), 7.94 (d, J = 8.5 Hz, 2H), 7.70 (t, J = 7.3 Hz, 9H), 7.44 (d, J = 8.5 Hz, 4H), 7.35 (d, J = 8.5 Hz, 2H), 7.29 (d, J = 8.5 Hz, 4H), 6.95−6.76 (m, 8H), 6.25 (d, J = 4.6 Hz, 2H), 5.80 (d, J = 16.0 Hz, 2H), 3.26 (s, 6H); ^{13}C NMR ($CDCl_3$, 75.3 MHz) 151.5, 150.8, 149.3, 146.8, 142.5, 135.1, 133.8, 125.8, 124.8, 123.8, 99.2, 52.7, 33.5, 29.6; MS (MALDI-TOF) m/z $[M]^+$ calcd for $C_{78}H_{53}N_9Zn$ 1179.37, found 1179.40;

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HRMS (MALDI-TOF) m/z $[M]^+$ calcd for $C_{78}H_{53}N_9Zn$ 1179.3715, found 1179.3752.

*PTP_M(OTEG)-2Py (**4**).* Following procedure A, 200 mg of **20** (142 μmol, 1.0 equiv) reacted with NaH (27 mg, 1.14 mmol, 8 equiv) and diethyl (4-pyridinyl)methyl phosphonate (130 mg, 0.57 mmol, 4 equiv) in dry THF (5 mL). Silica gel chromatography eluted with CH_2Cl_2/EtOH (99/1 to 95/5, v/v) followed by recrystallization from CH_2Cl_2/MeOH afforded the title compound with 99% yield (220 mg) as a dark blue powder: UV−vis in DMSO λ_{max} nm (ε mM^{-1} cm^{-1}) 451 (228.8), 582 (12.9), 632 (40.3); ^1H NMR ($CDCl_3$, 300 MHz) 9.67 (d, J = 4.2 Hz, 2H), 8.88 (d, J = 4.1 Hz, 2H), 8.78 (s, 4H), 8.04 (d, J = 7.7 Hz, 6H), 7.81 (d, J = 7.9 Hz, 2H), 7.25−7.15 (m, 16H), 6.98 (m, 4H), 6.75 (d, J = 16.1 Hz, 2H), 6.38 (m, 6H), 4.37 (m, 6H), 4.01 (m, 6H), 3.84 (m, 6H), 3.76 (m, 6H), 3.72 (m, 6H), 3.60 (m, 6H), 3.40 (s, 9H); ^{13}C NMR ($CDCl_3$, 75.3 MHz) 158.3, 151.9, 150.9, 150.2, 149.8, 147.2, 146.6, 144.6, 135.7, 135.5, 133.2, 132.9, 132.7, 132.6, 131.4, 130.7, 130.1, 128.1, 124.0, 123.5, 121.3, 119.9, 112.3, 98.5, 95.3, 94.1, 70.2−67.6, 59.1; MS (MALDI-TOF) m/z $[M]^+$ calcd for $C_{93}H_{89}N_7O_{12}Zn$ 1559.59, found 1559.59; HRMS (MALDI-TOF) (m/z) $[M]^+$ calcd for $C_{93}H_{89}N_7O_{12}Zn$ 1559.5861, found 1559.5862.

*PTP_M(DEGMan(OH)$_4$)$_3$-2Py (**5**).* A freshly prepared solution of sodium methanolate in dry methanol (4 mL, 0.1 M, 400 μmol, 24 equiv) was added to a solution of **31** (40 mg, 17 μmol, 1 equiv) in anhydrous DCM (4 mL). The solution was stirred at room temperature for 1 h. Then IWT ion-exchange resin was added, and the mixture was stirred carefully. After 30 min, the resin was filtered off and washed with a mixture of pyridine and water (1/1, v/v). Solvents were evaporated under reduced pressure. The reaction gave the title compound with a quantitative yield (30 mg) as a dark blue powder: UV−vis in DMSO λ_{max} nm (ε mM^{-1} cm^{-1}) 450 (159.2), 581 (5.8), 639 (29.8); ^1H NMR (pyridine-d_5, 300 MHz) 10.26 (d, J = 4.4 Hz, 2H), 9.32 (d, J = 4.4 Hz, 2H), 9.17 (s, 4H), 8.70 (d, J = 4.0 Hz, 4H), 8.26 (d, J = 7.9 Hz, 6H), 8.18 (d, J = 8.4 Hz, 2H), 7.72 (d, J = 8.4 Hz, 4H), 7.60 (d, J = 10.5 Hz, 2H), 7.50 (d, J = 5.5 Hz, 4H), 7.41 (d, J = 7.7 Hz, 8H), 7.34 (d, J = 8.4 Hz, 4H), 7.29−7.13 (m, 2H), 5.50 (s, 3H), 5.29 (br s, 9H), 4.66 (m, 12H), 4.41 (m, 12H), 4.20 (m, 3H), 4.14−4.06 (m, 3H), 4.02 (s, 6H), 3.90 (m, 9H); ^{13}C NMR (pyridine-d_5, 75.3 MHz) 171.2, 171.0, 159.6, 153.3, 153.2, 151.2, 148.1, 147.9, 145.6, 140.0, 133.8, 133.4, 133.2, 132.8, 131.6, 129.5, 126.4, 126.0, 125.5, 125.2, 121.7, 119.8, 113.7, 102.5, 100.1, 97.2, 94.9, 76.0, 74.6, 73.6, 72.6, 71.4, 70.5, 69.7, 68.6, 67.5, 65.3, 63.7, 62.3, 56.6; MS (MALDI-TOF) m/z $[M]^+$ calcd for $C_{102}H_{101}N_7O_{24}Zn$ 1871.62, found 1871.74, 1894.73 $[M + Na]^+$; HRMS (MALDI-TOF) (m/z) $[M]^+$ calcd for $C_{102}H_{101}N_7O_{24}Zn$ 1871.6189, found 1871.6199.

Photophysics. Fluorescence quantum yields (Φ_F) were measured according to Crosby's comparative method using Coumarin 153 in EtOH (Φ_F = 0.38) as reference.[52] One-photon singlet oxygen ($^1O_2$) generation was detected by its phosphorescence at 1270 nm through a PTI S/N 1565 monochromator, and the emission was monitored by a liquid nitrogen-cooled Ge-detector model (EO-817L, North Coast Scientific Co). Excitation occurred with a Xe-arc; the light was separated in a SPEX 1680, 0.22 μm double monochromator. The $^1O_2$ quantum yields (Φ_Δ) were calculated by a comparative method using tetraphenylporphyrin in DCM (Φ_Δ = 0.60) or Rose Bengal in DMF (Φ_Δ = 0.47) as a standard in the Crosby method. Characterization of the two-photon absorption properties of the dyes was obtained after two-photon-induced fluorescence (TPIF) measurements, with a femtosecond Ti:sapphire laser source delivering 100 fs pulses at 76 MHz repetition rate over the spectral range from 770 to 900 nm. The TPIF intensities of the samples were measured relative to a solution of Fluorescein, the ratio of the fluorescent signals enabling further determination of the 2PA cross section (δ) assuming equal one- and two-photon fluorescence quantum yields.[53]

Cell Lines and Cell Culture Conditions. The human retinoblastoma cell line Y79 and the human colorectal adenocarcinoma cell line HT-29 were obtained from the American Type Culture Collection. Y79 cells (cultured in suspension) and adherent HT-29 cells were grown in Dulbecco's modified Eagle's medium (DMEM) Glutamax supplemented with 20% and 10% fetal calf serum (FCS), respectively and antibiotics in humidified atmosphere under 5% CO_2 in air at 37 °C.

Dark Toxicity and Phototoxicity Assays. 5×10^5 Y79 cells and 5×10^4 HT-29 cells per well in 1 mL of appropriate culture medium were seeded into 24-well plates. After 5 h of incubation at 37 °C for Y79 and 24 h for HT-29, compounds **4** and **5**, in DMSO solution at 10^{-3} or 10^{-4} M, were added in the dark at a final concentration ranging from 0.75 to 7.5 μM in duplicate. Two plates of each cell line were realized, one for dark toxicity and the other for phototoxicity. Control cells received 7.5 μL of DMSO free of dye. After 24 h of incubation with compound at 37 °C in the dark, the cells were washed with phosphate buffered saline (PBS) and fresh medium free of drug was added. Illumination was performed for 27 min and 20 s (5 J/cm^2) through the bottom of the 24-well plates using a "light box" made of six Phillips TL 13W tubes (emission wavelength between 400 and 800 nm), leading to a final irradiance of 3 mW/cm^2. Plates were left to incubate at 37 °C in the dark for 3 days before evaluation of the cell viability by determination of mitochondrial activity using the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide (MTT, Sigma) assay. At the time of counting, 50 μL of a MTT (5 mg/mL) solution was added to each well. After 30 min of incubation and removal of the medium, formazan crystals were taken up with 600 μL of DMSO and absorbance at 562 nm was measured with a Fluostar microplate reader. Survival rate was expressed as ratio of the absorbance of treated and illuminated cells to 0.75% DMSO treated controls. IC_{50} (half maximal inhibitory concentration) values corresponding to the concentration of drug leading to 50% of survival, were calculated from the dose−response curves and are expressed in μM.

Cellular Uptake. Internalization of compounds **4** and **5** was measured by flow cytometry using a FACSCalibur four color instrument with CellQuest software. This FACSCalibur was equipped with a 488 nm argon laser with 530 nm (±15 nm, FL1-H), 585 nm (±21 nm, FL2-H) band-pass fluorescence filters and 670 nm (>670 nm, FL3-H) long-pass filter and a 635 nm red diode laser with a 661 nm band-pass filter (±8 nm, FL4-H).

For Y79 cells and HT-29 cells, 5×10^6 cells/well in 2 mL were seeded into 6-well plates with the appropriate culture medium (supplemented with 20% FCS for Y79 and 10% for HT-29). After 24 h of incubation, the compounds were added to a final concentration of 7.5 μM. Control cells were incubated with 15 μL of DMSO (0.75%). After 24 h of incubation in the dark at 37 °C, the cells were harvested. Y79 cells were centrifuged, washed and resuspended in 500 μL of PBS. HT-29 cells were trypsinized, centrifuged, washed and resuspended in 500 μL of PBS. Cells were analyzed by flow cytometry with the appropriate settings for each cell line. A least, fifty thousand cells were acquired initially and cells visualized on the basis of size (forward scatter) and granularity (side scatter). The debris and aggregates were excluded. Results are expressed as the percentage of compound-positive cells and the measurement of cellular uptake level using mean (MFI) of each detector. The ratio of MFI was calculated by dividing the value of treated cells by the one of control for each compound and each cell line. Finally, the means of three independent experiments were calculated.

■ ASSOCIATED CONTENT

Supporting Information

^1H and ^{13}C NMR spectra of all new compounds. This material is available free of charge via the Internet at http://pubs.acs.org.

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Notes

The authors declare no competing financial interest.

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