dye conjugates for use in such methods, for example, strategies that minimize or avoid photodegradation and improve the activity of the conjugate when used for PIT. Provided are methods and conjugates that meet such needs.

Summary

[0005] Provided in some examples is a method of manufacturing a phthalocyanine dye-conjugate. In some examples, the method includes contacting a targeting molecule, such as a macromolecule, with a phthalocyanine dye, which in some cases contains a reactive chemical group, under conditions to produce a conjugate containing the phthalocyanine dye linked to, such as covalently bound to, the targeting molecule. In some examples, the method includes formulating the conjugate in a pharmaceutically acceptable buffer. In some examples, before, during, and/or after preparation of the conjugate, the only light to which the dye and conjugate are exposed has a wavelength within a range from about 400 nm to about 650 nm. In some examples, before, during, and/or after preparation of the conjugate, such as during the contacting and/or formulating steps, the only light to which the dye and conjugate are exposed has an intensity of less than 500 lux.

[0005a] In one aspect, the present invention provides a method of manufacturing a phthalocyanine dye-targeting molecule conjugate, comprising:

a) contacting a targeting molecule with a phthalocyanine dye under conditions to produce a conjugate comprising the phthalocyanine dye linked to the targeting molecule; and

b) formulating the conjugate in a pharmaceutically acceptable buffer,

wherein in each of steps a) – b):

the only light to which the dye and conjugate are exposed has a wavelength within a range from about 400 nm to about 650 nm, or

the only light to which the dye and conjugate are exposed has an intensity of less than 500 lux.

[0006] In some examples, the method includes contacting a targeting molecule with a phthalocyanine dye at a molar ratio of dye to targeting molecule of from or from about 1:1 to 1000:1. In some examples, the dye includes a reactive chemical group, under conditions to produce a conjugate containing the phthalocyanine dye covalently linked to an attachment group of the targeting molecule. In some examples, the method includes formulating the conjugate in a pharmaceutically acceptable buffer to a concentration from or from about 0.01 mg/mL to 1000.0 mg/mL. In

some examples, before, during, and/or after preparation of the conjugate, such as during the contacting and/or formulating steps, the only light to which the dye and conjugate are exposed has an intensity of less than 500 lux.

[0007] In some examples, the conjugate is formulated to a concentration from or from about 0.01 mg/mL to about 200.0 mg/mL or from about 0.5 mg/mL to about 10.0 mg/mL. In some examples, the conjugate is formulated to a concentration from or from about 0.5 mg/mL to about 5.0 mg/mL.

[0008] In some examples, prior to the contacting step, the phthalocyanine dye is dissolved in a solvent under conditions in which the only light to which the dye is exposed has a wavelength within a range from about 400 nm to about 650 nm. In some examples, the only light to which the dye is exposed during or after being dissolved in solvent has an intensity of less than 500 lux. In some examples, the dye is dissolved in the solvent to a

concentration in a range from or from about 0.1 mg/mL to about 100 mg/mL. In some examples, the dye is dissolved in the solvent to a concentration from or from about 1 mg/mL to about 50 mg/mL. In some examples, the concentration of the phthalocyanine dye in the solvent is about 10 mg/mL. In some examples, the solvent is dimethylsulfoxide (DMSO) or DMF and water-based solvents.

[0009] In some examples, the formulating step includes concentrating the conjugate.

[0010] In some examples, the contacting step is carried out for at least 5 minutes, at least 15 minutes, at least 30 minutes, at least 60 minutes, at least 90 minutes, at least 120 minutes, at least 240 minutes, at least 360 minutes, at least 24 hours, at least 72 hours, or at least 120 hours. In some examples, the contacting step is carried out for 5 minutes to 150 hours, 5 minutes to 100 hours, 5 minutes to 48 hours, 5 minutes to 24 hours, 5 minutes to 6 hours, 5 minutes to 2 hours, 5 minutes to 90 minutes, 5 minutes to 60 minutes, 5 minutes to 30 minutes, 30 minutes to 150 hours, 30 minutes to 100 hours, 30 minutes to 48 hours, 30 minutes to 24 hours, 30 minutes to 6 hours, 30 minutes to 2 hours, 30 minutes to 90 minutes, 30 minutes to 60 minutes, 60 minutes to 150 hours, 60 minutes to 100 hours, 60 minutes to 48 hours, 60 minutes to 24 hours, 60 minutes to 6 hours, 60 minutes to 2 hours, 60 minutes to 90 minutes, 90 minutes to 150 hours, 90 minutes to 100 hours, 90 minutes to 48 hours, 90 minutes to 24 hours, 90 minutes to 6 hours, 90 minutes to 2 hours, 2 hours to 150 hours, 2 hours to 100 hours, 2 hours to 48 hours, 2 hours to 24 hours, 2 hours to 6 hours, 6 hours to 150 hours, 6 hours to 100 hours, 6 hours to 48 hours, 6 hours to 24 hours, 24 hours to 150 hours, 24 hours to 100 hours, 24 hours to 48 hours, 48 hours to 150 hours, 48 hours to 100 hours or 100 hours to 150 hours. In some examples, the contacting step is carried out at a temperature between or between about 4° C and about 37° C. In some examples, the contacting step is carried out at a temperature of about 25° C ± 2.0 °C, 25° C ± 1.0 °C or 25° C ± 0.3°C.

[0011] In some examples, the phthalocyanine dye is covalently or non-covalently linked to the targeting molecule. In some examples, the phthalocyanine dye contains a reactive chemical group and contacting the phthalocyanine dye and targeting molecule produces a conjugate comprising the phthalocyanine dye covalently bound to an attachment group of the targeting molecule.

[0012] In some examples, the method further includes quenching the conjugate, such as to remove unconjugated dye. In some examples, the only light to which the conjugate is exposed during the quenching step has a wavelength within a range from about 400 nm to

about 650 nm. In some examples, the only light to which the conjugate is exposed during the quenching step has an intensity of less than 500 lux.

[0013] In some examples, the formulating step includes ultrafiltration, diafiltration or dialysis. In some examples, the method further includes sterile filtration of the conjugate.

[0014] In some examples, the method includes packaging the conjugate, such as in one or more light-protected container. In some examples, during the packaging step the only light to which the conjugate is exposed has a wavelength within a range from about 400 nm to about 650 nm. In some examples, during the packaging step, the only light to which the dye and conjugate are exposed has an intensity of less than 500 lux.

[0015] In some examples, the methods include dissolving a phthalocyanine dye, which, in some cases containsa reactive chemical group, in a solvent to a concentration of about 0.1 -100 mg/mL. In some examples, the method further includes contacting a targeting molecule with the phthalocyanine dye at a molar ratio of dye to targeting molecule from 1:1 to 1000:1, under conditions to produce a conjugate containing the phthalocyanine dye linked to, *e.g.*, covalently bound to, the targeting molecule. In some examples, the method further includes formulating the conjugate in a pharmaceutically acceptable buffer to a concentration from or from about 0.01 to 1000.0 mg/mL. In some examples, the method further includes packaging the conjugate in one or more light-protected container. In some examples, before, during and/or after the steps of the method, such as the dissolving, contacting, formulating, and packaging steps, the only light to which the dye and conjugate are exposed has a wavelength within a range from about 400 nm to about 650 nm, or the only light to which the dye and conjugate are exposed has an intensity of less than 500 lux.

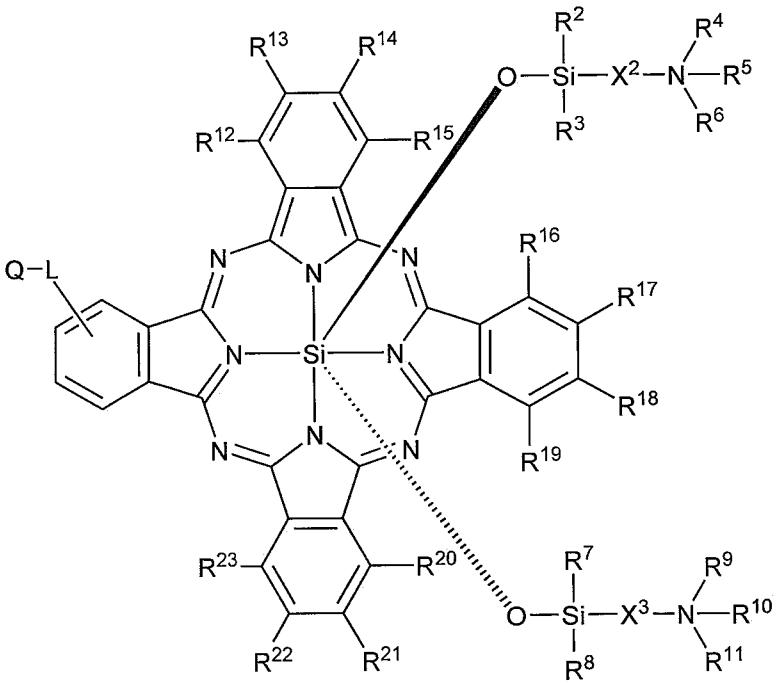
[0016] Provided in some examplesis a method of manufacturing a phthalocyanine dye-targeting molecule conjugate, comprising: a) dissolving a phthalocyanine dye in a solvent to a concentration of about 0.1 -100 mg/mL; b) contacting a targeting molecule with the phthalocyanine dye at a molar ratio of dye to targeting molecule from 1:1 to 1000:1 under conditions to produce a conjugate comprising the phthalocyanine dye linked to the targeting molecule; c) formulating the conjugate in a pharmaceutically acceptable buffer to a concentration from or from about 0.01 to about 200.0 mg/mL; and d) packaging the conjugate in one or more light-protected container, wherein in each of steps a) – d): the only light to which the dye and conjugate are exposed has a wavelength within a range from about 400 nm to about 650 nm, or the only light to which the dye and conjugate are exposed has an intensity of less than 500 lux.

[0017] In some of any such examples, the phthalocyanine dye is covalently or non-covalently linked to the targeting molecule. In some of any such examples, the phthalocyanine dye includes a reactive chemical group and contacting the phthalocyanine dye and targeting molecule produces a conjugate comprising the phthalocyanine dye covalently bound to an attachment group of the targeting molecule.

[0018] In some examples, during the steps of the provided methods, the total exposure of the dye and conjugate to any light is for no more than 5000 lux hours, no more than 2500 lux hours, no more than 1000 lux hours, no more than 500 lux hours, no more than 250 lux hours, no more than 100 lux hours or no more than 80 lux hours. In some examples, during the packaging step of the provided method, the total exposure of the conjugate to any light is for no more than 5000 lux hours, no more than 2500 lux hours, no more than 1000 lux hours, no more than 500 lux hours, no more than 250 lux hours, no more than 100 lux hours or no more than 80 lux hours.

[0019] In some examples, the dye has a maximum absorption wavelength from or from about 600 nm to about 850 nm. In some examples, the dye has a maximum absorption wavelength from or from about 650 nm to about 850 nm. In some examples, the dye has a maximum absorption wavelength from or from about 680 nm to about 850 nm.

[0020] In some examples, the phthalocyanine dye contains the formula:



, wherein:

L is a linker;

Q is a reactive group for attachment of the dye to the targeting molecule;

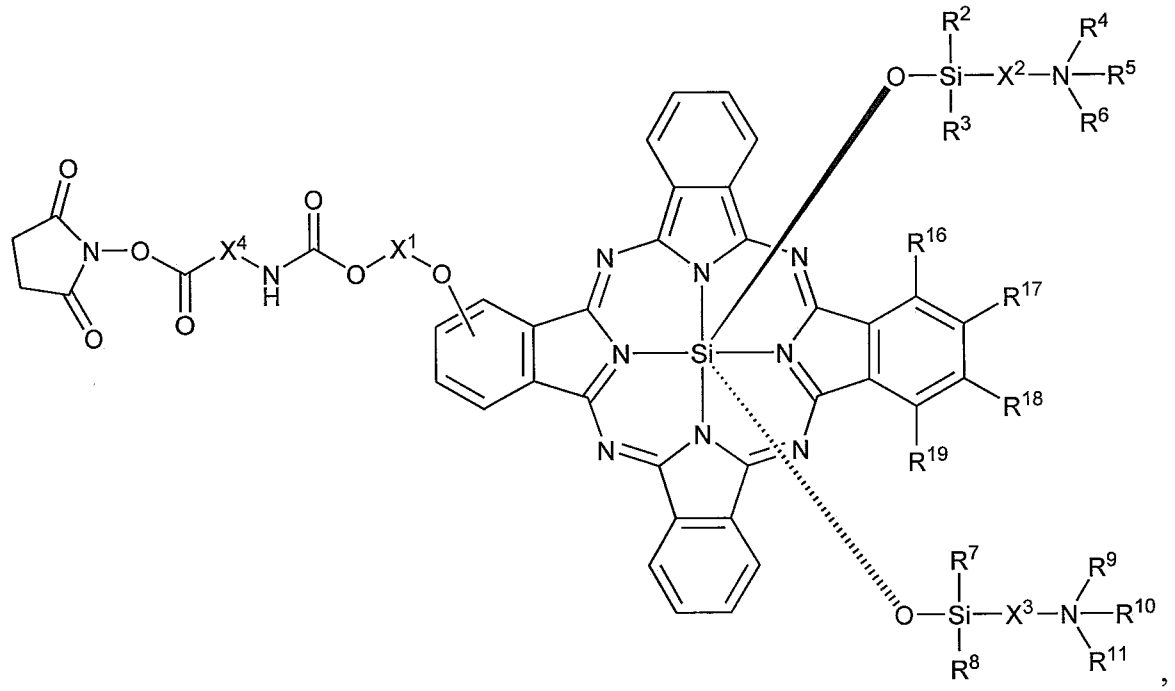
R^2, R^3, R^7, and R^8 are each independently selected from among optionally substituted alkyl and optionally substituted aryl;

R^4, R^5, R^6, R^9, R^{10}, and R^{11} are each independently selected from among hydrogen, optionally substituted alkyl, optionally substituted alkanoyl, optionally substituted alkoxycarbonyl, optionally substituted alkylcarbamoyl, and a chelating ligand, wherein at least one of R^4, R^5, R^6, R^9, R^{10}, and R^{11} comprises a water soluble group;

R^{12}, R^{13}, R^{14}, R^{15}, R^{16}, R^{17}, R^{18}, R^{19}, R^{20}, R^{21}, R^{22} and R^{23} are each independently selected from among hydrogen, halogen, optionally substituted alkylthio, optionally substituted alkylamino and optionally substituted alkoxy; and

X^2 and X^3 are each independently C_1–C_{10} alkylene, optionally interrupted by a heteroatom.

[0021] In some examples, the phthalocyanine dye includes the formula:



wherein:

X^1 and X^4 are each independently a C_1-C_{10} alkylene optionally interrupted by a heteroatom;

R^2, R^3, R^7, and R^8 are each independently selected from optionally substituted alkyl and optionally substituted aryl;

R^4, R^5, R^6, R^9, R^{10}, and R^{11} are each independently selected from among hydrogen, optionally substituted alkyl, optionally substituted alkanoyl, optionally substituted alkoxycarbonyl, optionally substituted alkylcarbamoyl, and a chelating ligand, wherein at least one of R^4, R^5, R^6, R^9, R^{10}, and R^{11} comprises a water soluble group; and

R^{16}, R^{17}, R^{18} and R^{19} are each independently selected from among hydrogen, halogen, optionally substituted alkylthio, optionally substituted alkylamino and optionally substituted alkoxy.

[0022] In some examples, the reactive group is selected from among an amine-reactive chemical group, a sulfhydryl-reactive chemical group, an activated ester, an acyl halide, an alkyl halide, an anhydride, a carboxylic acid, a carbodiimide, a carbonate, a carbamate, a haloacetamide, an isocyanate, an isothiocyanate, a maleimide, an NHS ester, a phosphoramidite, a platinum complex, a sulfonate ester and a thiocyanate. In some examples, the reactive chemical group is a sulfhydryl-reactive chemical group selected from among maleimides, haloacetyls and pyridyl disulfides. In some examples, the phthalocyanine dye is covalently bound to a lysine residue of the targeting molecule. In some examples, the reactive group is an amine-reactive chemical group. In some examples, the reactive chemical group is an amine-reactive chemical group that is an N-hydroxysuccinimide (NHS) ester.

[0023] In some examples, the phthalocyanine dye is IRDye 700DX-NHS (IR700-NHS).

[0024] In some examples, the targeting molecule binds to an antigen or protein directly or indirectly. For example, in some examples, the targeting molecule is a second binding molecule that binds to a first binding molecule, said first binding molecule being capable of binding to the antigen or protein. In some examples, the targeting molecule is a secondary antibody.

[0025] In some examples, the targeting molecule binds a cell surface target molecule on a surface of a cell or pathogen, such as a stem cell, a proliferating cell, a cancer cell, a cell in a hyperplasia, a tumor cell, an inflammatory cell, a neuron, a pathogen, or a pathogen infected cell. In some examples, the pathogen is selected from among viruses, bacteria, fungi, biofilms, and other prokaryote cell systems. In some examples, the cell is a cancer cell, a tumor cell, an inflammatory cell or, a neuron. In some examples, the cell is present in the microenvironment of a lesion associated with a disease or condition. In some examples, the lesion is a tumor and the cell is a cancer cell or a tumor cell. In some examples, the cell is a cancer stem cell or a circulating tumor cell.

[0026] In some such examples, the inflammatory cell is a leukocyte such as a neutrophil, an eosinophil, a basophil, a lymphocyte, or a monocyte. In some examples, the targeting molecule is a neuron, such as a peripheral nervous system neuron or a central nervous system neuron. In some examples, the neuron is a nociceptor such as a

nociceptor, mechanical nociceptor, chemical nociceptor or polymodal nociceptor. In some examples, the targeting molecule binds to a pathogen, such as a virus, bacterium, fungus, biofilm, or other prokaryote cell system. In some examples, the pathogen is a gram-negative or gram-positive bacterium.

[0027] In some examples, the cell surface target molecule includes an antigen, a polypeptide, a lipid, or a carbohydrate, or a combination thereof.

[0028] In some examples, the cell surface target molecule is selected from among cell membrane phospholipids, prokaryotic peptidoglycans, bacterial cell envelop proteins, viral capsid proteins, ACTHR, endothelial cell Anxa-1, aminopetidase N, anti-IL-6R, alpha-4-integrin, alpha-5-beta-3 integrin, alpha-5-beta-5 integrin, alpha-fetoprotein (AFP), ANPA, ANPB, APA, APN, APP, 1AR, 2AR, AT1, B1, B2, BAGE1, BAGE2, B-cell receptor BB1, BB2, BB4, calcitonin receptor, cancer antigen 125 (CA 125), CCK1, CCK2, CD5, CD10, CD11a, CD13, CD14, CD19, CD20, CD22, CD25, CD30, CD33, CD38, CD45, CD52, CD56, CD68, CD90, CD133, CD7, CD15, CD34, CD44, CD206, CD271, CEA (CarcinoEmbryonic Antigen), CGRP, chemokine receptors, cell-surface annexin-1, cell-surface plectin-1, Cripto-1, CRLR, CXCR2, CXCR4, DCC, DLL3, E2 glycoprotein, EGFR, EGFRvIII, EMR1, Endosialin, EP2, EP4, EpCAM, EphA2, ET receptors, Fibronectin, Fibronectin ED-B, FGFR, frizzled receptors, GAGE1, GAGE2, GAGE3, GAGE4, GAGE5, GAGE6, GLP-1 receptor, G-protein coupled receptors of the Family A (Rhodopsin-like), G-protein coupled receptors of the Family B (Secretin receptor-like) like), G-protein coupled receptors of the Family C (Metabotropic Glutamate Receptor-like), GD2, GP100, GP120, Glypican-3, hemagglutinin, Heparin sulfates, HER1, HER2, HER3, HER4, HMFG, HPV 16/18 and E6/E7 antigens, hTERT, an interleukin receptor (*e.g.* IL-2R, IL11-R, IL-13R) ITGAM, Kalikrien-9, Lewis Y, LH receptor, LHRH-R, LPA1, MAC-1, MAGE 1, MAGE 2, MAGE 3, MAGE 4, MART1, MC1R, Mesothelin, MUC1, MUC16, Neu (cell-surface Nucleolin), Neprilysin, Neuropilin-1, Neuropilin-2, NG2, NK1, NK2, NK3, NMB-R, Notch-1, NY-ESO-1, OT-R, mutant p53, p97 melanoma antigen, NTR2, NTR3, p32 (p32/gC1q-R/HABP1), p75, PAC1, PAR1, Patched (PTCH), PDGFR, PDFG receptors, PDT, Protease-cleaved collagen IV, proteinase 3, prohibitin, protein tyrosine kinase 7, PSA, PSMA, purinergic P2X family (*e.g.* P2X1-5), mutant Ras, RAMP1, RAMP2, RAMP3 patched, RET receptor, plexins, smoothened, sst1, sst2A, sst2B, sst3, sst4, sst5, substance P, TEMs, T-cell CD3 Receptor, TAG72, TGFBR1, TGFBR2, Tie-1, Tie-2, Trk-A, Trk-B, Trk-C, TR1, TRPA, TRPC, TRPV, TRPM, TRPML, TRPP (*e.g.* TRPV1-6, TRPA1, TRPC1-7, TRPM1-8, TRPP1-5, TRPML1-3), TSH receptor, VEGF receptors (VEGFR1 or Flt-1, VEGFR2 or

FLK-1/KDR, and VEGF-3 or FLT-4), voltage-gated ion channels, VPAC1, VPAC2, Wilms tumor 1, Y1, Y2, Y4 and Y5.

[0029] In some examples, the cell surface target molecule is selected from among HER1/EGFR, HER2/ERBB2, CD20, CD25 (IL-2Rα receptor), CD33, CD52, CD133, CD206, CEA, CEACAM1, CEACAM3, CEACAM5, CEACAM6, cancer antigen 125 (CA125), alpha-fetoprotein (AFP), Lewis Y, TAG72, Caprin-1, mesothelin, PDGF receptor, PD-1, PD-L1, CTLA-4, IL-2 receptor, vascular endothelial growth factor (VEGF), CD30, EpCAM, EphA2, Glypican-3, gpA33, mucins, CAIX, PSMA, folate-binding protein, gangliosides (such as GD2, GD3, GM1 and GM2), VEGF receptor (VEGFR),VEGFR2, VEGF-A, integrin αVβ3, integrin α5β1, ERBB3, MET, IGF1R, EPHA3, TRAILR1, TRAILR2, RANKL, FAP, tenascin, AFP, BCR complex, CD3, CD18, CD44, CTLA-4, gp72, HLA-DR 10 β, HLA-DR antigen, IgE, MUC-1, nuC242, PEM antigen, metalloproteinases, Ephrin receptor, Ephrin ligands, HGF receptor, CXCR4, CXCR4, Bombesin receptor, SK-1antigen, Bcr-abl, RET, MET, TRKB, TIE2, ALK, ROS, EML4-ALK, ROS1, BRAFV600E, SRC, c-KIT, PDGFR, mTOR, TSC1, TSC2, BTK, KIT, BRCA, CDK 4/6, JAK1, JAK2, BRAF, FLT-3, MEK1, MEK2, and SMO. In some examples, the cell surface target molecule is HER1/EGFR, HER2, PD-L1, CD25, EpCAM, EphA2, CD206, CD20, CD44, CD133, Mesothelin, Glypican-3, or carcinoembryonic antigen (CEA).

[0030] In some examples, at least part of the targeting molecule is or is a combination of a protein, a glycoprotein, an antibody, an antibody fragment, an antigen, an antigen binding fragment, a peptide, a polypeptide, a tissue homing peptide, a small molecule, a polymeric synthetic molecule, a polymeric nanoparticle, a liposome, an enzyme substrate, a hormone, a neurotransmitter, a cell metabolite, a viral particle, a viral capsid, a viral nanoparticle, a bacterial particle, a marker, a cell, a hapten, an avidin, a streptavidin, a monomeric streptavidin, a biotin, a carbohydrate, an oligosaccharide, a polysaccharide, a nucleic acid, a deoxy nucleic acid, a fragment of DNA, a fragment of RNA, an aptamer, nucleotide triphosphates, acyclo terminator triphosphates, or PNA.

[0031] In some examples, the targeting molecule is a tissue-specific homing peptide. In some examples, the homing peptide has the sequence of amino acids as set forth in any of SEQ ID NOS: 1-52.

[0032] In some examples, the targeting molecule is an RGD polypeptide, an iRGD polypeptide, a Lyp-1 polypeptide, a cripto-1 binding polypeptide, a somatostatin receptor binding polypeptide, a prohibitin binding polypeptide, a NGR polypeptide, an iNGR

polypeptide, or an activatable cell penetrating peptide (ACPP) comprised of a polycationic cell penetrating peptide (CPP) connected via a cleavable linker to a neutralizing polyanion.

[0033] In some examples, the ACPP comprises the structure: A-X1-B-, wherein B is a peptide portion of about 5 to about 20 basic amino acid residues, which is suitable for cellular uptake; A is a peptide portion of about 2 to about 20 acidic amino acid residues, which when linked with portion B is effective to inhibit or prevent cellular uptake of portion B; X1 is a cleavable linker of about 2 to about 100 atoms; and one or more of L-Y is linked to the C-terminus of peptide portion B.

[0034] In some examples, the targeting molecule is selected from among adrenocorticotropic hormone (ACTH), angiotensin II, atrial natriuretic factor (ANF), bombesin, bradykinin, brain derived neurotropihic factor (BDNF), bone morphogenetic protein 2 (BMP-2), bone morphogenetic protein 6 (BMP-6), bone morphogenetic protein 7 (BMP-7), calcitonin, cardiotrophin 1 (BMP-2), CD22, CD40, cholecystokinin (CCK), ciliary neurotrophic factor (CNTF), CCL1-CCL28, CXCL1-CXCL17, XCL1, XCL2, CX3CL1, cripto 1 binding peptide, vascular endothelial cell growth factor (VEGF), epidermal growth factor (EGF), endothelin 1, endothelin 1/3, FAS-ligand, fibroblast growth factor 1 (FGF-1), fibroblast growth factor 2 (FGF-2), fibroblast growth factor 4 (FGF-4), fibroblast growth factor 5 (FGF-5), fibroblast growth factor 6 (FGF-6), fibroblast growth factor 1 (FGF-7), fibroblast growth factor 1 (FGF-10), Flt-3, gastrin, gastrin releasing peptide (GRP), granulocyte colony-stimulating factor (G-CSF), granulocyte macrophage stimulating factor (GM-CSF), glucagon like peptide (GLP-1), hepatocyte growth factor (HGF), interferon alpha (IFN-a), interferon beta (IFN-b), interferon gamma (IFNg), insulin-like growth factor 1(IGF-1), insulin-like growth factor 2 (IGF-2), interleukin 1 (IL-1), interleukin 2 (IL-2), interleukin 3 (IL-3), interleukin 4 (IL-4), interleukin 5 (IL-5), interleukin 6 (IL-6), interleukin 7 (IL-7), interleukin 8 (IL-8), interleukin 9 (IL-9), interleukin 10 (IL-10), interleukin 11 (IL-11), interleukin 12 (IL-12), interleukin 13 (IL-13), interleukin 15 (IL-15), interleukin 17 (IL-17), interleukin 19 (IL-19), luteinizing hormone (LH), luteinizing-releasing hormone (LHRH), macrophage colony-stimulating factor (M-CSF), monocyte chemotactic protein 1 (MCP-1), macrophage inflammatory protein 3a (MIP-3a), macrophage inflammatory protein 3b (MIP-3b), nerve growth factor (NGF), neuromedin B, neurotrophin 3 (NT-3), neurotrophin 4 (NT-4), neurotensin, neuropeptide Y, oxytocin, pituitary adenylate cyclase activating peptide (PACAP), platelet derived growth factor AA (PDGF-AA), platelet derived growth factor AB (PDGF-AB), platelet derived growth factor BB (PDGF-BB), platelet derived growth factor CC (PDGF-CC), platelet derived growth factor DD (PDGF-DD), netrin-1 (NTN1), netrin-2

(NTN2), netrin-4 (NTN4), netrin-G1 (NTNG1) and netrin-G2 (NTNG2), ephrin A1 (EFNA1), ephrin A2 (EFNA2), ephrin A3 (EFNA3), ephrin A4 (EFNA4), ephrin A5 (EFNA5), semaphorin 3A (SEMA3A), semaphorin 3B (SEMA3B), semaphorin 3C (SEMA3C), semaphorin 3D (SEMA3D), semaphorin 3F (SEMA3F), semaphorin 3G (SEMA3G), semaphorin 4A (SEMA4A), semaphorin 4B (SEMA4B), semaphorin 4C (SEMA4C), semaphorin 4D (SEMA4D), semaphorin 4F (SEMA4F), semaphorin 4G (SEMA4G), semaphorin 5A (SEMA5A), semaphorin 5B (SEMA5B), semaphorin 6A (SEMA6A), semaphorin 6B (SEMA6B), semaphorin 6D (SEMA6D), semaphorin 7A (SEMA7A), SLIT1, SLIT2, SLIT3, SLIT and NTRK-like family, member 1 (SLITRK1), SLIT and NTRK-like family, member 2 (SLITRK2), SLIT and NTRK-like family, member 3 (SLITRK3), SLIT and NTRK-like family, member 4 (SLITRK4), SLIT and NTRK-like family, member 5 (SLITRK5), SLIT and NTRK-like family, member 6 (SLITRK6), prostaglandin E2 (PGE2), RANTES, Somatostatin-14, Somatostatin-28, stem cell factor (SCF), stromal cell derived factor 1 (SDF-1), substance P, thyroid stimulating hormone (TSH), transforming growth factor alpha (TGF-α), transforming growth factor beta (TGF-b), tumor necrosis factor alpha (TNF-α), thrombin, vasoactive intestinal peptide (VIP), Wntl, Wnt2, Wnt2b/13, Wnt3, Wnt3a, Wnt4, Wnt5a, Wnt5b, Wnt6, Wnt7a, Wnt7b, Wnt7c, Wnt8, Wnt8a, Wnt8b, Wnt8c, Wntl0a, Wntl0b, Wnt11, Wnt14, Wnt15, or Wnt16, Sonic hedgehog, Desert hedgehog, and Indian hedgehog.

[0035] In some examples, the targeting molecule is an antibody or an antibody fragment.

[0036] In some examples, the antibody is cetuximab, panitumumab, zalutumumab, nimotuzumab, trastuzumab, Ado-trastuzumab emtansine, Tositumomab (Bexxar ®), Rituximab (Rituxan, Mabthera), Ibritumomab tiuxetan (Zevalin), Daclizumab (Zenapax), Gemtuzumab (Mylotarg), Alemtuzumab, CEA-scan Fab fragment, OC125 monoclonal antibody, ab75705, B72.3, Bevacizumab (Avastin ®), Afatinib, Axitinib, Bosutinib, Cabozantinib, Ceritinib, Crizotinib, Dabrafenib, Dasatinib, Erlotinib, Everolimus, Ibrutinib, Imatinib, Lapatinib, Lenvatinib, Nilotinib, Olaparib, Palbociclib, Pazopanib, Pertuzumab, Ramucirumab, Regorafenib, Ruxolitinib, Sorafenib, Sunitinib, Temsirolimus, Trametinib, Vandetanib, Vemurafenib, Vismodegib, Basiliximab, Ipilimumab, Nivolumab, pembrolizumab, MPDL3280A, Pidilizumab (CT-011), AMP-224, MSB001078C, or MEDI4736, or is an antigen-binding fragment thereof. In some examples, the antibody binds to a cell surface target molecule, such as HER1/EGFR, HER2, PD-L1, CD25, EpCAM, EphA2, CD206, CD20, CD44, CD133, Mesothelin, Glypican-3, or carcinoembryonic antigen

(CEA). In some examples, the antibody is cetuximab, Panitumumab, Trastuzumab, BMS-935559, MEDI4736, MPDL3280A or MSB0010718C or is an antigen-binding fragment thereof.

[0037] In some examples, the dye-targeting molecule conjugate is cetuximab-IR700, Panitumumab-IR700, Trastuzumab-IR700, BMS-935559-IR700, MEDI4736-IR700, MPDL3280A-IR700 or MSB0010718C-IR700.

[0038] In some examples, the targeting molecule is contacted with the phthalocyanine dye at a molar ratio of dye to targeting molecule from 1:1 to 100:1 or 1:1 to 10:1. In some examples, the molar ratio of dye to targeting molecule is at least or at least about 4:1 or is at least or at least about 10:1. In some examples, the conjugate includes from or from about 1 to about 1000 phthalocyanine dye molecules per targeting molecule, from or from about 1 to about 10 phthalocyanine dye molecules per targeting molecule or from or from about 2 to about 5 phthalocyanine dye molecules per targeting molecule.

[0039] In some examples, the conjugate is formulated to a concentration that is from or from about 1.0 to about 5.0 mg/mL, such as in a pharmaceutically acceptable buffer. In some examples, the pharmaceutically acceptable buffer is phosphate buffered saline. In some examples, the pharmacologically acceptable buffer has a pH from or from about pH 6.0 to about pH 8.0. In some examples, the conjugate is stable for greater than 3 months with greater than 90% of the conjugate present as a main monomer component. In some examples, the conjugate is stable if it retains greater than or greater than about 30%, 40%, 50%, 60%, 70%, 80%, 90% or more of its potency, activity or purity for greater than 3 months compared to the conjugate prior to the storage for the time. In some examples, the conjugate is stable if greater than 90% of the conjugate is present as a main monomer component. In some examples, the pharmacologically acceptable buffer has a pH from or from about pH 6.8 to about pH 7.4.

[0040] In some examples, the only light to which the dye and conjugate are exposed has a wavelength within a range from about 425 nm to about 575 nm. In some examples, the only light to which the dye and conjugate are exposed has an intensity of less than 200 lux.

[0041] In some examples, the container comprising the conjugate protects from transmission of light having a wavelength from or from about 250 nm to about 800 nm, from about 250 nm to about 450 nm, from about 400 nm to about 800 nm, from about 450 nm to about 650 nm, or from about 600 nm to about 720 nm. In some examples, container protects from transmission of light such that the percentage of light transmission by the

container is less than 50%, less than 40%, less than 30%, less than 20%, less than 10% or less than 5%. In some examples, the container is green, blue, amber, translucent, opaque, or is wrapped in an opaque foil. In some examples, the container is green, blue, amber, translucent, opaque, or is covered by material with light transmission of less than 50%, less than 40%, less than 30%, less than 20%, less than 10% or less than 5%. In some examplesof the methods provided herein, the container is selected from among a vial, a tube, a syringe, a bag, a pouch, and a box.

[0042] In some examples, the light-protected container is a first light-protected container and the method further includes packing the first light-protected container into a second light-protected container. In some examples, the second container protects from transmission of light having a wavelength from or from about 250 nm to about 800 nm, from about 250 nm to about 450 nm, from about 400 nm to about 800 nm, from about 450 nm to about 650 nm, or from about 600 nm to about 720 nm. In some examples, the second container protects from transmission of light such that the percentage of light transmission is less than 50%, less than 40%, less than 30%, less than 20%, less than 10% or less than 5%. In some examples, the second container is green, blue, amber, translucent, opaque, or is covered by a material with light transmission of less than 50%, less than 40%, less than 30%, less than 20%, less than 10% or less than 5%.In some examples, the second container is selected from among a vial, a tube, a syringe, a bag, a pouch, and a box.

[0043] In some examples, the method provided herein further includes packaging the second container into a third light-protected container. In some examples, the third container protects from transmission of light having a wavelength from or from about 250 nm to about 800 nm, from about 250 nm to about 450 nm, from about 400 nm to about 800 nm, from about 450 nm to about 650 nm, or from about 600 nm to about 720 nm. In some examples, the third container protects from transmission of light such that the percentage of light transmission is less than 50%, less than 40%, less than 30%, less than 20%, less than 10% or less than 5%. In some examples, the third container is green, blue, amber, translucent, opaque, or is covered by a material with light transmission of less than 50%, less than 40%, less than 30%, less than 20%, less than 10% or less than 5%. In some examples, the third container is selected from among a vial, a tube, a syringe, a bag, a pouch, and a box.

[0044] In some examples, the amount of the conjugate produced by the method is greater than or greater than about 1 gram, greater than or greater than about 2 grams, greater than or greater than about 3 grams, greater than or greater than about 4 grams, greater than or

greater than about 5 grams or greater than or greater than about 10 grams. In some examples, the conjugate is produced using good manufacturing practice (GMP).

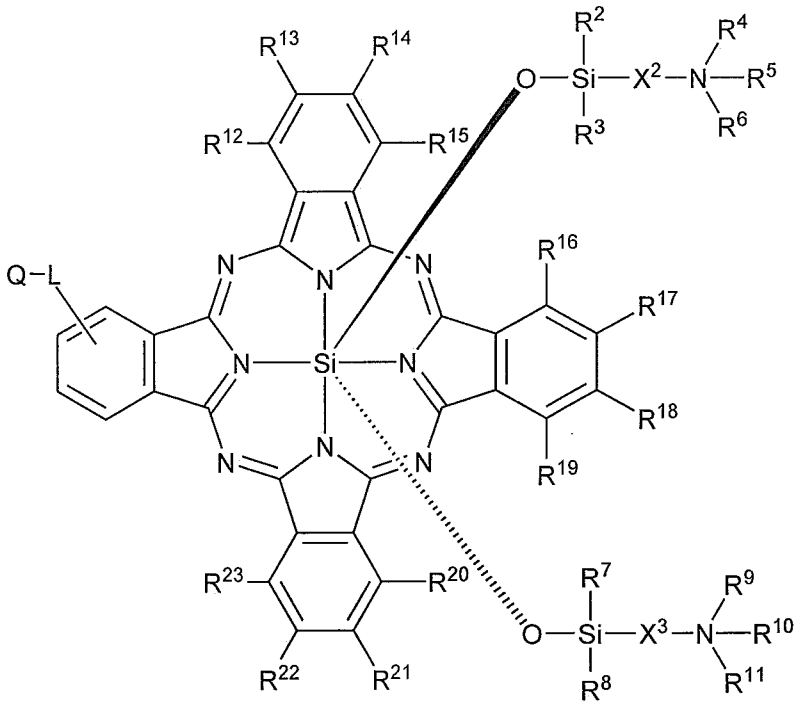
[0045] Provided in some examplesis a conjugate produced, formulated or packaged by the method described herein. In some examples, the conjugate is stable for greater than three months, such as with greater than 90% of the conjugate present as a main monomer component.

[0046] Provided in some examplesis a stable conjugate containing a phthalocyanine dye linked to a targeting molecule. In some examples, the stable conjugate is stable for greater than three months, such as with greater than 90% of the conjugate present as a main monomer component.

[0046a] In another aspect, the present invention provides a stable conjugate, comprising the phthalocyanine dye-targeting molecule conjugate manufactured by the method described herein, wherein the conjugate is stable for greater than three months or greater than 6 months or greater than 12 months.

[0047] In some examples, the conjugate is stable if it retains greater than or greater than about 30%, 40%, 50%, 60%, 70%, 80%, 90% or more of its potency, activity or purity for greater than 3 months compared to the conjugate prior to the storage for the time. In some examples, the conjugate is stable if greater than 90% of the conjugate or stable conjugate is present as a main monomer component. In some examples, the conjugate is stable if greater than 95% of the conjugate or stable conjugate is present as a main monomer component. In some examples, the conjugate or stable conjugate is stable for greater than 6 months or greater than 12 months. In some examples, the conjugate or stable conjugate is stable at a temperature of less than 30 °C.

[0048] In some examples, the phthalocyanine dye contained in the stable conjugate includes the formula:



, wherein:

L is a linker;

Q is a reactive group for attachment of the dye to the targeting molecule;

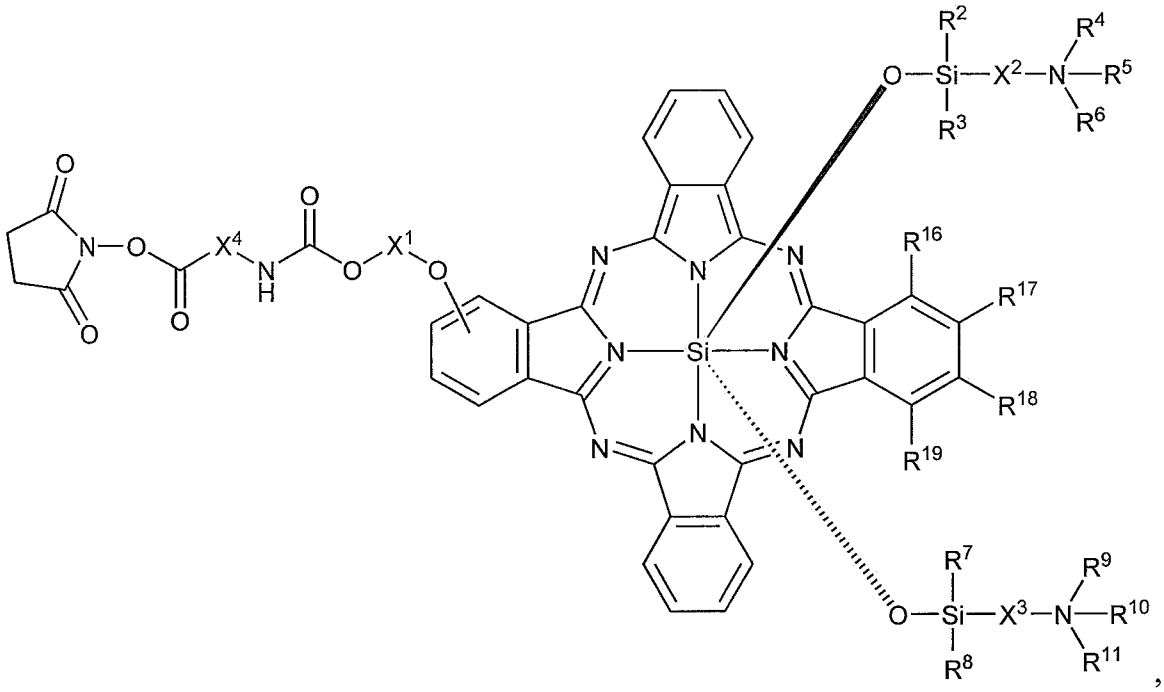
R^2, R^3, R^7, and R^8 are each independently selected from among optionally substituted alkyl and optionally substituted aryl;

R^4, R^5, R^6, R^9, R^{10}, and R^{11} are each independently selected from among hydrogen, optionally substituted alkyl, optionally substituted alkanoyl, optionally substituted alkoxycarbonyl, optionally substituted alkylcarbamoyl, and a chelating ligand, wherein at least one of R^4, R^5, R^6, R^9, R^{10}, and R^{11} comprises a water soluble group;

R^{12}, R^{13}, R^{14}, R^{15}, R^{16}, R^{17}, R^{18}, R^{19}, R^{20}, R^{21}, R^{22} and R^{23} are each independently selected from among hydrogen, halogen, optionally substituted alkylthio, optionally substituted alkylamino and optionally substituted alkoxy; and

X^2 and X^3 are each independently C_1–C_{10} alkylene, optionally interrupted by a heteroatom.

[0049] In some examples, the stable conjugate containing the phthalocyanine dye includes the formula:



wherein:

X^1 and X^4 are each independently a C_1-C_{10} alkylene optionally interrupted by a heteroatom;

R^2, R^3, R^7, and R^8 are each independently selected from optionally substituted alkyl and optionally substituted aryl;

R^4, R^5, R^6, R^9, R^{10}, and R^{11} are each independently selected from among hydrogen, optionally substituted alkyl, optionally substituted alkanoyl, optionally substituted alkoxycarbonyl, optionally substituted alkylcarbamoyl, and a chelating ligand, wherein at least one of R^4, R^5, R^6, R^9, R^{10}, and R^{11} comprises a water soluble group; and

R^{16}, R^{17}, R^{18} and R^{19} are each independently selected from among hydrogen, halogen, optionally substituted alkylthio, optionally substituted alkylamino and optionally substituted alkoxy

[0050] In some examples, the dye contained in the stable conjugate has a maximum absorption wavelength from or from about 600 nm to about 850 nm, from or from about 650 nm to about 850 nm, or from or from about 680 nm to about 850 nm.

[0051] In some examples, the dye contained in the stable conjugate is IRDye 700DX (IR700).

[0052] In some examples, the targeting molecule contained in the stable conjugate binds to a cell surface target molecule on a surface of a cell or pathogen, such as a proliferating cell, a cancer cell, a cell in hyperplasia, a tumor cell, an inflammatory cell, a neuron, or a pathogen. In some examples, the cell is a stem cell, a proliferating cell, a

cell in a hyperplasia, or a pathogen infected cell. In some examples, the pathogen is selected from among viruses, bacteria, fungi, biofilms, and other prokaryote cell systems. In some examples, the inflammatory cell is a leukocyte, such as a neutrophil, an eosinophil, a basophil, a lymphocyte, or a monocyte. In some examples, the targeting molecule is a neuron, such as a peripheral nervous system neuron or a central nervous system neuron. In some examples, the neuron is a nociceptor such as a nociceptor, mechanical nociceptor, chemical nociceptor or polymodal nociceptor. In some examples, the targeting molecule binds to a pathogen, such as a virus, bacterium, fungus, biofilm, or other prokaryote cell system. In some examples, the pathogen is a gram-negative or gram-positive bacterium.

[0053] In some examples, the cell surface target molecule bound by the targeting molecule contained in the stable conjugate includes an antigen, a polypeptide, a lipid, or a carbohydrate, or a combination thereof.

[0054] In some examples, the cell surface target molecule is selected from among cell membrane phospholipids, prokaryotic peptidoglycans, bacterial cell envelop proteins, viral capsid proteins, ACTHR, endothelial cell Anxa-1, aminopetidase N, anti-IL-6R, alpha-4-integrin, alpha-5-beta-3 integrin, alpha-5-beta-5 integrin, alpha-fetoprotein (AFP), ANPA, ANPB, APA, APN, APP, 1AR, 2AR, AT1, B1, B2, BAGE1, BAGE2, B-cell receptor BB1, BB2, BB4, calcitonin receptor, cancer antigen 125 (CA 125), CCK1, CCK2, CD5, CD10, CD11a, CD13, CD14, CD19, CD20, CD22, CD25, CD30, CD33, CD38, CD45, CD52, CD56, CD68, CD90, CD133, CD7, CD15, CD34, CD44, CD206, CD271, CEA (CarcinoEmbryonic Antigen), CGRP, chemokine receptors, cell-surface annexin-1, cell-surface plectin-1, Cripto-1, CRLR, CXCR2, CXCR4, DCC, DLL3, E2 glycoprotein, EGFR, EGFRvIII, EMR1, Endosialin, EP2, EP4, EpCAM, EphA2, ET receptors, Fibronectin, Fibronectin ED-B, FGFR, frizzled receptors, GAGE1, GAGE2, GAGE3, GAGE4, GAGE5, GAGE6, GLP-1 receptor, G-protein coupled receptors of the Family A (Rhodopsin-like), G-protein coupled receptors of the Family B (Secretin receptor-like) like), G-protein coupled receptors of the Family C (Metabotropic Glutamate Receptor-like), GD2, GP100, GP120, Glypican-3, hemagglutinin, Heparin sulfates, HER1, HER2, HER3, HER4, HMFG, HPV 16/18 and E6/E7 antigens, hTERT, an interleukin receptor (*e.g.* IL-2R, IL11-R, IL-13R), ITGAM, Kalikrien-9, Lewis Y, LH receptor, LHRH-R, LPA1, MAC-1, MAGE 1, MAGE 2, MAGE 3, MAGE 4, MART1, MC1R, Mesothelin, MUC1, MUC16, Neu (cell-surface Nucleolin), Neprilysin, Neuropilin-1, Neuropilin-2, NG2, NK1, NK2, NK3, NMB-R, Notch-1, NY-ESO-1, OT-R, mutant p53, p97 melanoma antigen, NTR2, NTR3, p32 (p32/gC1q-R/HABP1), p75, PAC1, PAR1, Patched (PTCH), PDGFR, PDFG receptors, PDT, Protease-

cleaved collagen IV, proteinase 3, prohibitin, protein tyrosine kinase 7, PSA, PSMA, purinergic P2X family (*e.g.* P2X1-5), mutant Ras, RAMP1, RAMP2, RAMP3 patched, RET receptor, plexins, smoothened, sst1, sst2A, sst2B, sst3, sst4, sst5, substance P, TEMs, T-cell CD3 Receptor, TAG72, TGFBR1, TGFBR2, Tie-1, Tie-2, Trk-A, Trk-B, Trk-C, TR1, TRPA, TRPC, TRPV, TRPM, TRPML, TRPP (*e.g.* TRPV1-6, TRPA1, TRPC1-7, TRPM1-8, TRPP1-5, TRPML1-3), TSH receptor, VEGF receptors (VEGFR1 or Flt-1, VEGFR2 or FLK-1/KDR, and VEGF-3 or FLT-4), voltage-gated ion channels, VPAC1, VPAC2, Wilms tumor 1, Y1, Y2, Y4, and Y5.

[0055] In some examples, the cell surface target molecule is selected from among HER1/EGFR, HER2/ERBB2, CD20, CD25 (IL-2Rα receptor), CD33, CD52, CD133, CD206, CEA, CEACAM1, CEACAM3, CEACAM5, CEACAM6, cancer antigen 125 (CA125), alpha-fetoprotein (AFP), Lewis Y, TAG72, Caprin-1, mesothelin, PDGF receptor, PD-1, PD-L1, CTLA-4, IL-2 receptor, vascular endothelial growth factor (VEGF), CD30, EpCAM, EphA2, Glypican-3, gpA33, mucins, CAIX, PSMA, folate-binding protein, gangliosides (such as GD2, GD3, GM1 and GM2), VEGF receptor (VEGFR),VEGFR2, VEGF-A, integrin αVβ3, integrin α5β1, ERBB3, MET, IGF1R, EPHA3, TRAILR1, TRAILR2, RANKL, FAP, tenascin, AFP, BCR complex, CD3, CD18, CD44, CTLA-4, gp72, HLA-DR 10 β, HLA-DR antigen, IgE, MUC-1, nuC242, PEM antigen, metalloproteinases, Ephrin receptor, Ephrin ligands, HGF receptor, CXCR4, CXCR4, Bombesin receptor, SK-1antigen, Bcr-abl, RET, MET, TRKB, TIE2, ALK, ROS, EML4-ALK, ROS1, BRAFV600E, SRC, c-KIT, PDGFR, mTOR, TSC1, TSC2, BTK, KIT, BRCA, CDK 4/6, JAK1, JAK2, BRAF, FLT-3, MEK1, MEK2, and SMO. In some examples, the cell surface target molecule is HER1/EGFR, HER2, PD-L1, CD25, EpCAM, EphA2, CD206, CD20, CD44, CD133, Mesothelin, Glypican-3, or carcinoembryonic antigen (CEA).

[0056] In some examples, at least part of the targeting molecule is or is a combination of a protein, a glycoprotein, an antibody, an antibody fragment, an antigen, an antigen binding fragment, a peptide, a polypeptide, a tissue homing peptide, a small molecule, a polymeric synthetic molecule, a polymeric nanoparticle, a liposome, an enzyme substrate, a hormone, a neurotransmitter, a cell metabolite, a viral particle, a viral capsid, a viral nanoparticle, a bacterial particle, a marker, a cell, a hapten, an avidin, a streptavidin, a monomeric streptavidin, a biotin, a carbohydrate, an oligosaccharide, a polysaccharide, a nucleic acid, a deoxy nucleic acid, a fragment of DNA, a fragment of RNA, an aptamer, nucleotide triphosphates, acyclo terminator triphosphates, or PNA.

[0057] In some examples, the targeting molecule is a tissue-specific homing peptide. In some examples, the homing peptide has the sequence of amino acids as set forth in any of SEQ ID NOS: 1-52.

[0058] In some examples, the targeting molecule is an RGD polypeptide, an iRGD polypeptide, a Lyp-1 polypeptide, a cripto-1 binding polypeptide, a somatostatin receptor binding polypeptide, a prohibitin binding polypeptide, a NGR polypeptide, an iNGR polypeptide, or an activatable cell penetrating peptide (ACPP) comprised of a polycationic cell penetrating peptide (CPP) connected via a cleavable linker to a neutralizing polyanion.

[0059] In some examples, the ACPP comprises the structure: A-X1-B-, wherein B is a peptide portion of about 5 to about 20 basic amino acid residues, which is suitable for cellular uptake; A is a peptide portion of about 2 to about 20 acidic amino acid residues, which when linked with portion B is effective to inhibit or prevent cellular uptake of portion B; X1 is a cleavable linker of about 2 to about 100 atoms; and one or more of L-Y is linked to the C-terminus of peptide portion B.

[0060] In some examples, the targeting molecule is selected from among adrenocorticotropic hormone (ACTH), angiotensin II, atrial natriuretic factor (ANF), bombesin, bradykinin, brain derived neurotropihic factor (BDNF), bone morphogenetic protein 2 (BMP-2), bone morphogenetic protein 6 (BMP-6), bone morphogenetic protein 7 (BMP-7), calcitonin, cardiotrophin 1 (BMP-2), CD22, CD40, cholecystokinin (CCK), ciliary neurotrophic factor (CNTF), CCL1-CCL28, CXCL1-CXCL17, XCL1, XCL2, CX3CL1, cripto 1 binding peptide, vascular endothelial cell growth factor (VEGF), epidermal growth factor (EGF), endothelin 1, endothelin 1/3, FAS-ligand, fibroblast growth factor 1 (FGF-1), fibroblast growth factor 2 (FGF-2), fibroblast growth factor 4 (FGF-4), fibroblast growth factor 5 (FGF-5), fibroblast growth factor 6 (FGF-6), fibroblast growth factor 1 (FGF-7), fibroblast growth factor 1 (FGF-10), Flt-3, gastrin, gastrin releasing peptide (GRP), granulocyte colony-stimulating factor (G-CSF), granulocyte macrophage stimulating factor (GM-CSF), glucagon like peptide (GLP-1), hepatocyte growth factor (HGF), interferon alpha (IFN-a), interferon beta (IFN-b), interferon gamma (IFNg), insulin-like growth factor 1(IGF-1), insulin-like growth factor 2 (IGF-2), interleukin 1 (IL-1), interleukin 2 (IL-2), interleukin 3 (IL-3), interleukin 4 (IL-4), interleukin 5 (IL-5), interleukin 6 (IL-6), interleukin 7 (IL-7), interleukin 8 (IL-8), interleukin 9 (IL-9), interleukin 10 (IL-10), interleukin 11 (IL-11), interleukin 12 (IL-12), interleukin 13 (IL-13), interleukin 15 (IL-15), interleukin 17 (IL-17), interleukin 19 (IL-19), luteinizing hormone (LH), luteinizing-releasing hormone (LHRH), macrophage colony-stimulating factor (M-CSF), monocyte chemotactic protein 1 (MCP-1),

macrophage inflammatory protein 3a (MIP-3a), macrophage inflammatory protein 3b (MIP-3b), nerve growth factor (NGF), neuromedin B, neurotrophin 3 (NT-3), neurotrophin 4 (NT-4), neurotensin, neuropeptide Y, oxytocin, pituitary adenylate cyclase activating peptide (PACAP), platelet derived growth factor AA (PDGF-AA), platelet derived growth factor AB (PDGF-AB), platelet derived growth factor BB (PDGF-BB), platelet derived growth factor CC (PDGF-CC), platelet derived growth factor DD (PDGF-DD), netrin-1 (NTN1), netrin-2 (NTN2), netrin-4 (NTN4), netrin-G1 (NTNG1) and netrin-G2 (NTNG2), ephrin A1 (EFNA1), ephrin A2 (EFNA2), ephrin A3 (EFNA3), ephrin A4 (EFNA4), ephrin A5 (EFNA5), semaphorin 3A (SEMA3A), semaphorin 3B (SEMA3B), semaphorin 3C (SEMA3C), semaphorin 3D (SEMA3D), semaphorin 3F (SEMA3F), semaphorin 3G (SEMA3G), semaphorin 4A (SEMA4A), semaphorin 4B (SEMA4B), semaphorin 4C (SEMA4C), semaphorin 4D (SEMA4D), semaphorin 4F (SEMA4F), semaphorin 4G (SEMA4G), semaphorin 5A (SEMA5A), semaphorin 5B (SEMA5B), semaphorin 6A (SEMA6A), semaphorin 6B (SEMA6B), semaphorin 6D (SEMA6D), semaphorin 7A (SEMA7A), SLIT1, SLIT2, SLIT3, SLIT and NTRK-like family, member 1 (SLITRK1), SLIT and NTRK-like family, member 2 (SLITRK2), SLIT and NTRK-like family, member 3 (SLITRK3), SLIT and NTRK-like family, member 4 (SLITRK4), SLIT and NTRK-like family, member 5 (SLITRK5), SLIT and NTRK-like family, member 6 (SLITRK6), prostaglandin E2 (PGE2), RANTES, Somatostatin-14, Somatostatin-28, stem cell factor (SCF), stromal cell derived factor 1 (SDF-1), substance P, thyroid stimulating hormone (TSH), transforming growth factor alpha (TGF-α), transforming growth factor beta (TGF-b), tumor necrosis factor alpha (TNF-α), thrombin, vasoactive intestinal peptide (VIP), Wntl, Wnt2, Wnt2b/13, Wnt3, Wnt3a, Wnt4, Wnt5a, Wnt5b, Wnt6, Wnt7a, Wnt7b, Wnt7c, Wnt8, Wnt8a, Wnt8b, Wnt8c, Wntl0a, Wntl0b, Wnt11, Wnt14, Wnt15, or Wnt16, Sonic hedgehog, Desert hedgehog, and Indian hedgehog.

[0061] In some examples, the targeting molecule is an antibody or an antibody fragment.

[0062] In some examples, the antibody is cetuximab, panitumumab, zalutumumab, nimotuzumab, trastuzumab, Ado-trastuzumab emtansine, Tositumomab (Bexxar ®), Rituximab (Rituxan, Mabthera), Ibritumomab tiuxetan (Zevalin), Daclizumab (Zenapax), Gemtuzumab (Mylotarg), Alemtuzumab, CEA-scan Fab fragment, OC125 monoclonal antibody, ab75705, B72.3, Bevacizumab (Avastin ®), Afatinib, Axitinib, Bosutinib, Cabozantinib, Ceritinib, Crizotinib, Dabrafenib, Dasatinib, Erlotinib, Everolimus, Ibrutinib, Imatinib, Lapatinib, Lenvatinib, Nilotinib, Olaparib, Palbociclib, Pazopanib, Pertuzumab,

Ramucirumab, Regorafenib, Ruxolitinib, Sorafenib, Sunitinib, Temsirolimus, Trametinib, Vandetanib, Vemurafenib, Vismodegib, Basiliximab, Ipilimumab, Nivolumab, pembrolizumab, lambrolizumab, MPDL3280A, Pidilizumab (CT-011), MSB001078C, BMS-935559 or MEDI4736, AMP-224, or is an antigen-binding fragment thereof. In some examples, the antibody binds to a cell surface target molecule, such as HER1/EGFR, HER2, PD-L1, CD25, EpCAM, EphA2, CD206, CD20, CD44, CD133, Mesothelin, Glypican-3, or carcinoembryonic antigen (CEA). In some examples, the antibody is cetuximab, Panitumumab, Trastuzumab, BMS-935559, MEDI4736, MPDL3280A or MSB0010718C or is an antigen-binding fragment thereof.

[0063] In some examples, the stable dye-targeting molecule conjugate is cetuximab-IR700, Panitumumab-IR700, Trastuzumab-IR700, BMS-935559-IR700, MEDI4736-IR700, MPDL3280A-IR700 or MSB0010718C-IR700.

[0064] In some examples, the stable conjugate includes from or from about 1 to about 1000 phthalocyanine dye molecules per targeting molecule, from or from about 1 to about 10 or from or from about 2 to about 5 phthalocyanine dye molecules per targeting molecule.

[0065] Provided in some examplesis a composition containing the conjugate or stable conjugate.

[0066] Provided in some examplesis a pharmaceutical composition containing the conjugate or stable conjugate and a pharmaceutically acceptable excipient.

[0066a] In yet another aspect, the present invention provides a pharmaceutical composition comprising the stable conjugate described herein and a pharmaceutically acceptable excipient.

[0067] In some examples, the composition is formulated in phosphate buffered saline. In some examples, composition has a pH of greater than 6.0.

[0068] Provided in some examplesis a pharmaceutical composition containing a phthalocyanine dye linked to a targeting molecule and a pharmaceutically acceptable excipient. In some such examples, the composition has a pH of greater than 6.0 and the conjugate in the composition is stable for greater than three months, such as with greater than 90% of the conjugate present as a main monomer component. In some such examples, the conjugate in the composition is stable if it retains greater than or greater than about 30%, 40%, 50%, 60%, 70%, 80%, 90% or more of its potency, activity or purity for greater than 3 months compared to the conjugate prior to the storage for the time. In some such examples, the conjugate in the composition is stable if greater than 90% of the conjugate is present as a main monomer component. In some examples, the pH of the composition is greater than 6.0 or is from or from about pH 6.0 to about 8.0, inclusive.

[0069] In some examples, the concentration of the conjugate in the composition is from or from about 0.01 mg/mL to about 200 mg/mL. In some examples, the concentration of the conjugate in the composition is from or from about 0.5 mg/mL to about 10 mg/mL. In some examples, the concentration of the conjugate in the composition is from or from about 1.0 to about 5.0 mg/mL. In some examples, the concentration of the conjugate in the composition is from or from about 1.8 to about 2.1 mg/mL.

[0070] In some examples, the volume of the composition is from or from about 0.5 mL to about 100 mL, from or from about 1 mL to about 50 mL or from or from about 1 mL to about 10 mL.

[0071] Provided in some examplesis a container comprising the conjugate or stable conjugate. In some examples, the container protects from transmission of light having a wavelength from or from about 500 nm to 725 nm or 650 nm to 725 nm. In some examples, the container protects from transmission of light such that the percentage of light transmission by the container is less than 50%, less than 40%, less than 30%, less than 20%, less than 10% or less than 5%. In some examples, the container is green, blue, amber, translucent, opaque, or is covered by a material with light transmission of less than 50%, less than 40%, less than 30%, less than 20%, less than 10% or less than 5%. In some examples, the container is wrapped in an opaque foil.

[0072] Provided in some examplesis a packaging system for protecting a phthalocyanine dye-targeting molecule conjugate from light. In some examples, the packaging system includes an internal packaging material comprising the container described herein. In some examples, the internal packaging material has a light transmittance of no more than 5%. In some examples, an external packaging material contains the internal packaging material. In some examples, the external packaging material has a light transmittance of no more than 5%. In some examples, the internal packaging material includes an opaque foil.

[0072a] In yet another aspect, the present invention provides a packaging system comprising the stable conjugate described herein, and further comprising a container, wherein the container protects from transmission of light such that the percentage of transmission of light is less than 50%.

[0073] Provided in some examplesis a packaging system for protecting a phthalocyanine dye-targeting molecule conjugate from light that includes a first container,

such as any container provided herein, and a second container comprising the first container, wherein the second container protects from transmission of light having a wavelength from or from about 250 nm to about 800 nm, from about 250 nm to about 450 nm, from about 400 nm to about 800 nm, from about 450 nm to about 650 nm, or from about 600 nm to about 720 nm. In some examples, the second container protects from transmission of light such that the percentage of light transmission is less than 50%, less than 40%, less than 30%, less than

20%, less than 10% or less than 5%. In some examples, the second container is green, blue, amber, translucent, opaque, or is covered by a material with light transmission of less than 50%, less than 40%, less than 30%, less than 20%, less than 10% or less than 5%. In some examples, the first and second container are independently selected from among a vial, a tube, a syringe, a bag, a pouch, and a box.

[0074] In some examples, any of the provided packaging system further includes a third container comprising the second container, wherein the third container protects from transmission of light having a wavelength from or from about 250 nm to about 800 nm, from about 250 nm to about 450 nm, from about 400 nm to about 800 nm, from about 450 nm to about 650 nm, or from about 600 nm to about 720 nm. In some examples, the third container protects from transmission of light such that the percentage of light transmission is less than 50%, less than 40%, less than 30%, less than 20%, less than 10% or less than 5%. In some examples, the third container is green, blue, amber, translucent, opaque, or is covered by a material with light transmission of less than 50%, less than 40%, less than 30%, less than 20%, less than 10% or less than 5%. In some examples, the third container is selected from among a vial, a tube, a syringe, a bag, a pouch, and a box.

[0075] Provided in some examplesis a kit that includes any of the containers described herein or any of the packaging systems described herein; a light-protected cover capable of covering a device capable of administering a composition comprising a phthalocyanine dye-targeting molecule conjugate; and optionally instructions for use. In some examples, the administration device is an intravenous infusion bag. In some examples, the light-protected cover protects from transmission of light having a wavelength from or from about 250 nm to about 800 nm, from about 250 nm to about 450 nm, from about 400 nm to about 800 nm, from about 450 nm to about 650 nm, or from about 600 nm to about 720 nm. In some examples, the light-protected cover protects from transmission of light such that the percentage of light transmission is less than 50%, less than 40%, less than 30%, less than 20%, less than 10% or less than 5%. In some examples, the light-protected cover is green, blue, amber, translucent, opaque, or is covered by a material with light transmission of less than 50%, less than 40%, less than 30%, less than 20%, less than 10% or less than 5%.

[0076] Provided in some examplesis a method of preparing a composition comprising a phthalocyanine-dye conjugate for administration that includes: unpacking one or more of any of the containers described herein or one or more of any of the packaging system described herein that includes any of the containers described herein; and transferring

the composition present in the one or more containers into a device capable of administering the composition to a subject, wherein the only light to which the composition is exposed has a wavelength within a range from about 400 nm to about 650 nm, or the only light to which the composition is exposed has an intensity of less than 500 lux. In some examples, the only light in which the composition is exposed has an intensity of less than 200 lux or less than 100 lux. In some examples, the method provided is performed in a biosafety cabinet, biosafety hood or a sterile environment. In some examples, the one or more containers together comprise a therapeutically effective dose of the phthalocyanine-dye conjugate. In some examples, the one or more containers include at least or about at least or 2, 4, 6, 8, 10, 12, 18 or 24 containers. In some examples, the provided method is carried out for no more than 1 hour, no more than 30 minutes or no more than 15 minutes; or the total exposure of the composition to any light during the method is no more than 500 lux hours, no more than 250 lux hours, no more than 100 lux hours, no more than 50 lux hours or no more than 25 lux hours.

[0077] In some examples, the administration device is an intravenous infusion bag. In some examples, the administration device comprises a light-protected cover capable of covering the device. In some examples, the light-protected cover protects from transmission of light having a wavelength from or from about 250 nm to about 800 nm, from about 250 nm to about 450 nm, from about 400 nm to about 800 nm, from about 450 nm to about 650 nm, or from about 600 nm to about 720 nm.

[0078] In some examples, the light-protected cover protects from transmission of light such that the percentage of light transmission is less than 50%, less than 40%, less than 30%, less than 20%, less than 10% or less than 5%. In some examples, the light-protected cover is green, blue, amber, translucent, opaque, or is covered by a material with light transmission of less than 50%, less than 40%, less than 30%, less than 20%, less than 10% or less than 5%.

[0079] Provided in some examplesis a light-protected device that includes the composition prepared using the methods provided herein.

[0080] Provided in some examplesis a method of removing unwanted cells or pathogens in a subject, that includes: (a) administering a composition comprising a phthalocyanine-dye conjugate from any of the light-protected device provided herein to a subject, wherein prior to and during the administration step the composition is not exposed to an intensity of environmental light greater than 500 lux; and (b) irradiating the unwanted

cells or pathogens at a wavelength of 660 to 740 nm at a dose of at least 1 J cm^{-2} or 1 J/cm of fiber length thereby removing the unwanted cell in the subject.

[0081] Provided in some examplesis a method of removing unwanted cells or pathogens in a subject that includes: a) administering to a subject a therapeutically effective amount of any of the conjugates or compositions described herein, wherein prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux; and b) irradiating the unwanted cells or pathogens at a wavelength of 660 to 740 nm at a dose of at least 1 J cm^{-2} or 1 J/cm of fiber length thereby removing the unwanted cell in the subject.

[0082] Provided in some examplesis a method of removing unwanted cells or pathogens in a subject that includes: a) administering to a subject a therapeutically effective amount of a conjugate comprising IRDye 700DX (IR700) linked to a targeting molecule capable of binding an unwanted cell or pathogen, wherein prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux; and b) irradiating the unwanted cells or pathogens at a wavelength of 600 to 800 nm at a dose of at least 1 J cm^{-2} or 1 J/cm of fiber length thereby removing the unwanted cells or pathogens in the subject.

[0083] Provided in some examplesis a method of removing unwanted cells or pathogens in a subject that includes: a) administering to a subject a therapeutically effective amount of a first binding molecule capable of binding an unwanted cell or pathogen; b) administering to the subject a conjugate molecule comprising IRDye 700DX (IR700) linked to a targeting molecule, wherein the targeting molecule is a second binding molecule that is capable of binding to the first binding molecule; and c) irradiating the unwanted cells or pathogens at a wavelength of 600 to 800 nm at a dose of at least 1 J cm^{-2} or 1 J/cm of fiber length thereby removing the unwanted cells or pathogens in the subject.

[0084] Provided in some examplesis a method of removing unwanted cells or pathogens in a sample, that includes: (a) administering a composition comprising a phthalocyanine-dye conjugate from any of the light-protected device provided herein to a sample, wherein prior to and during the administration step the composition is not exposed to an intensity of environmental light greater than 500 lux; and (b) irradiating the unwanted cells or pathogens at a wavelength of 660 to 740 nm at a dose of at least 1 J cm^{-2} or 1 J/cm of fiber length thereby removing the unwanted cell in the sample.

[0085] Provided in some examplesis a method of removing unwanted cells or pathogens in a sample that includes: a) administering to a sample a therapeutically effective amount of any of the conjugates or compositions described herein, wherein prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux; and b) irradiating the unwanted cells or pathogens at a wavelength of 660 to 740 nm at a dose of at least 1 J cm^{-2} or 1 J/cm of fiber length thereby removing the unwanted cell in the sample.

[0086] Provided in some examplesis a method of removing unwanted cells or pathogens in a sample that includes: a) administering to a sample a therapeutically effective amount of a conjugate comprising IRDye 700DX (IR700) linked to a targeting molecule capable of binding an unwanted cell or pathogen, wherein prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux; and b) irradiating the unwanted cells or pathogens at a wavelength of 600 to 800 nm at a dose of at least 1 J cm^{-2} or 1 J/cm of fiber length thereby removing the unwanted cells or pathogens in the sample.

[0087] Provided in some examplesis a method of removing unwanted cells or pathogens in a sample that includes: a) administering to a sample a therapeutically effective amount of a first binding molecule capable of binding an unwanted cell or pathogen; b) administering to the sample a conjugate molecule comprising IRDye 700DX (IR700) linked to a targeting molecule, wherein the targeting molecule is a second binding molecule that is capable of binding to the first binding molecule; and c) irradiating the unwanted cells or pathogens at a wavelength of 600 to 800 nm at a dose of at least 1 J cm^{-2} or 1 J/cm of fiber length thereby removing the unwanted cells or pathogens in the sample.

[0088] In some examples, the method is performed *in vitro* or *ex vivo*. In some examples, the method is performed using an extracorporeal device.

[0089] In some examplesof the methods provided herein, the first binding molecule is administered to the subject prior to the conjugate or the first binding molecule and conjugate are administered simultaneously to the subject. In some examples, the targeting molecule is a secondary antibody. In some examples, prior to and during the administration of the conjugate, the conjugate is not exposed to an intensity of environmental light greater than 500 lux.

[0090] In some examples, the cell is a stem cell, a proliferating cell, a cell in a hyperplasia, an inflammatory cell, a negative regulatory immune cell, which optionally is a T cell, a pathogen infected cell, a neuron, a fat cell or adipocyte. In some examples, the cell

is a cancer cell or a tumor cell. In some examples, the cell is associated with, causes or contributes to the etiology of a disease or condition. In some examples, the disease of condition is a tumor or cancer, an infection, an inflammatory disease or condition, or a neuronal disease or condition. In some examples, the cell is a neuron and the disease or condition is a neurological disorder, which optionally is pain; the cell is a fat cell or adipocyte and the disease or condition involves excess fat; the cell is a pathogen infected cell and the disease or condition is an infection; the cell is a pathogen and the disease or condition is an infection; the cell is an inflammatory cell and the disease or condition is an inflammatory disease; the cell is a an immune cell, which optionally is a regulatory T cell, and the disease or condition is a tumor or cancer; or the cell is a tumor or cancer cell and the disease or condition is a tumor or a cancer.

[0091] In some examples, the cell is present in the microenvironment of a lesion associated with a disease or condition or is in a hyperplasia. In some examples, the lesion is a tumor and the disease or condition is a tumor or cancer. In some examples, the method treats the disease or condition.

[0092] Provided in some examplesis a method of removing a pathogen infected cell in a subject that includes: a) administering to a subject a therapeutically effective amount of a conjugate molecule comprising IRDye 700DX (IR700) linked to a targeting molecule, wherein the targeting molecule is capable of binding to the pathogen infected cell directly or indirectly; and b) irradiating the pathogen infected cell at a wavelength of 600 to 800 nm at a dose of at least 1 J cm^{-2} or 1 J/cm of fiber length thereby removing the pathogen infected cell in the subject. In some examples, the pathogen is a virus, bacterium, fungus, biofilm, or other prokaryote cell system. In some examples, prior to and during the administration of the conjugate, the conjugate is not exposed to an intensity of environmental light greater than 500 lux.

[0093] Provided in some examplesis a method of removing a pathogen infected cell in a sample that includes: a) administering to a sample a therapeutically effective amount of a conjugate molecule comprising IRDye 700DX (IR700) linked to a targeting molecule, wherein the targeting molecule is capable of binding to the pathogen infected cell directly or indirectly; and b) irradiating the pathogen infected cell at a wavelength of 600 to 800 nm at a dose of at least 1 J cm^{-2} or 1 J/cm of fiber length thereby removing the pathogen infected cell in the sample. In some examples, the pathogen is a virus, bacterium, fungus, biofilm, or other prokaryote cell system. In some examples, prior to and during the administration of

the conjugate, the conjugate is not exposed to an intensity of environmental light greater than 500 lux.

[0094] In some examples, the method is performed *in vitro* or *ex vivo*. In some examples, the *ex vivo* irradiation is method is performed using an extracorporeal device. Provided in some examplesis a method of treating hyperplasia or a tumor in a subject. In some examples, the method includes administering to the subject a therapeutically effective amount of the conjugate or stable conjugate or composition, where prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux. In some examples, the method further includes irradiating the hyperplasia or the tumor at a wavelength of 660 to 740 nm at a dose of at least 1 J cm^{-2} or 1 J/cm of fiber length, thereby treating the tumor in the subject.

[0094a] In yet another aspect, the present invention provides use of a phthalocyanine dye-targeting molecule conjugate manufactured according to the method described herein for the preparation of a medicament for use in a treatment regimen for treating a hyperplasia or a tumor in a subject, the treatment regimen comprising:

a) administering to the subject a therapeutically effective amount of the phthalocyanine dye-targeting molecule conjugate, wherein prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux; and

b) irradiating the hyperplasia or the tumor at a wavelength of 660 to 740 nm at a dose of at least 1 J cm or 1 J/cm of fiber length thereby treating the tumor in the subject

[0095] Provided in some examplesis a method of treating hyperplasia or a tumor in a subject. In some such examples, the method includes administering to the subject a therapeutically effective amount of a conjugate containing IRDye 700DX (IR700) linked to a targeting molecule. In some examples, the conjugate is targeted to the hyperplasia or the tumor and prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux. In some examples, the method further includes irradiating the tumor at a wavelength of 600 to 800 nm at a dose of at least 1 J cm^{-2} or 1 J/cm of fiber length thereby treating the tumor in the subject.

[0095a] In yet another aspect, the present invention provides use of the stable conjugate described herein for the preparation of a medicament for use in a treatment regimen for treating a hyperplasia or a tumor in a subject, the treatment regimen comprising:

a) administering to the subject a therapeutically effective amount of the stable conjugate, wherein prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux; and

b) irradiating the hyperplasia or the tumor at a wavelength of 660 to 740 nm at a dose of at least 1 J cm or 1 J/cm of fiber length thereby treating the tumor in the subject.

[0096] Provided in some examplesis a method of treating hyperplasia or a tumor in a sample. In some examples, the method includes administering to the sample a therapeutically effective amount of the conjugate or stable conjugate or composition, where prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux. In some examples, the method further includes irradiating the hyperplasia or the tumor at a wavelength of 660 to 740 nm at a dose of at least 1 J cm^{-2} or 1 J/cm of fiber length, thereby treating the tumor in the sample.

[0096a] In yet another aspect, the present invention provides a pharmaceutical composition described herein for use in a treatment regimen for treating a hyperplasia or a tumor in a subject, the treatment regimen comprising:

a) administering to the subject a therapeutically effective amount of the phthalocyanine dye-targeting molecule conjugate, wherein prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux; and

b) irradiating the hyperplasia or the tumor at a wavelength of 660 to 740 nm at a dose of at least 1 J cm or 1 J/cm of fiber length thereby treating the tumor in the subject.

[0097] Provided in some examplesis a method of treating hyperplasia or a tumor in a sample. In some such examples, the method includes administering to the sample a therapeutically effective amount of a conjugate containing IRDye 700DX (IR700) linked to a targeting molecule. In some examples, the conjugate is targeted to the hyperplasia or the tumor and prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux. In some examples, the method further includes irradiating the tumor at a wavelength of 600 to 800 nm at a dose of at least 1 J cm^{-2} or 1 J/cm of fiber length thereby treating the tumor in the sample.

[0097a] In yet another aspect, the present invention provides the pharmaceutical composition described herein for use in a treatment regimen for treating a hyperplasia or a tumor in a subject, the treatment regimen comprising:

a) administering to the subject a therapeutically effective amount of the stable conjugate, wherein prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux; and

b) irradiating the hyperplasia or the tumor at a wavelength of 660 to 740 nm at a dose of at least 1 J cm or 1 J/cm of fiber length thereby treating the tumor in the subject.

[0097b] In yet another aspect, the present invention provides a method of removing unwanted cells or pathogens in a sample comprising:

a) administering to a sample, the sample comprising unwanted cells or pathogens, a therapeutically effective amount of the conjugate manufactured according to the method described herein, wherein prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux; and

b) irradiating the unwanted cells or pathogens at a wavelength of 660 to 740 nm at a dose of at least 1 J cm or 1 J/cm of fiber length thereby removing the unwanted cells or pathogens in the sample, optionally wherein the method is performed *in vitro* or *ex vivo*.

[0097c] In yet another aspect, the present invention provides use of a phthalocyanine dye-targeting molecule conjugate manufactured according to the method described herein for the preparation of a medicament for use in a treatment regimen for removing unwanted cells or pathogens in a subject, wherein the unwanted cells or pathogens are associated with a disease or condition in the subject and the treatment regimen comprising:

a) administering to the subject a therapeutically effective amount of the conjugate, wherein prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux; and

b) irradiating the unwanted cells or pathogens at a wavelength of 660 to 740 nm at a dose of at least 1 J cm or 1 J/cm of fiber length thereby removing the unwanted cells or pathogens in the subject and treating the disease or condition in the subject.

[0097d] In yet another aspect, the present invention provides a pharmaceutical composition described herein for use in a treatment regimen for removing unwanted cells or pathogens in a subject, wherein the unwanted cells or pathogens are associated with a disease or condition in the subject and the treatment regimen comprising:

a) administering to the subject a therapeutically effective amount of the conjugate, wherein prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux; and

b) irradiating the unwanted cells or pathogens at a wavelength of 660 to 740 nm at a dose of at least 1 J cm or 1 J/cm of fiber length thereby removing the unwanted cells or pathogens in the subject and treating the disease or condition in the subject.

[0098] In some examples, the method is for treating a tumor, wherein the targeting molecule of the conjugate targets the conjugate to the tumor or a microenvironment of the tumor. In some examples, irradiating the tumor is at a wavelength of 600 to 800 nm and at a dose of at least 1 J cm^{-2} or 1 J/cm of fiber length, thereby treating the tumor, e.g., the tumor in the subject or in the sample.

[0099] In some examples, the targeting molecule is an antibody, an antigen binding fragment, a protein, a glycoprotein, a peptide, a polypeptide, a virus, a viral capsid, or a viral particle. In some examples, the targeting molecule is an antibody or an antibody fragment.

[0100] In some examples, administration is performed under fluorescent lighting or LED lighting and in the absence of direct or indirect sunlight.

[0101] In some examples, any exposure of the conjugate to light less than 500 lux is for less than 20 minutes, less than 15 minutes, less than 10 minutes or less than 5 minutes. In some examples, the exposure of the conjugate to any light is light with an intensity that is not greater than 50 lux.

[0102] In some examples, the tumor is a cancer. In some examples, the cancer is a cancer located at the head and neck, breast, liver, colon, ovary, prostate, pancreas, brain, cervix, bone, skin, eye, bladder, stomach, esophagus, peritoneum, or lung. In some examples, the cancer is a cancer of the blood.

[0103] In some examples, the conjugate is targeted to a protein expressed in the tumor. In some examples, the conjugate is targeted to a protein expressed on the surface of a cell present in the tumor microenvironment. In some examples, the cell is a tumor cell, an immune cell or a cancer stem cell.

[0104] In some examples, the protein expressed in the tumor is ACTHR, endothelial cell Anxa-1, aminopetidase N, anti-IL-6R, alpha-4-integrin, alpha-5-beta-3 integrin, alpha-5-beta-5 integrin, alpha-fetoprotein (AFP), ANPA, ANPB, APA, APN, APP, 1AR, 2AR, AT1, B1, B2, BAGE1, BAGE2, B-cell receptor BB1, BB2, BB4, calcitonin receptor, cancer antigen 125 (CA 125), CCK1, CCK2, CD5, CD10, CD11a, CD13, CD14,

CD19, CD20, CD22, CD25, CD30, CD33, CD38, CD45, CD52, CD56, CD68, CD90, CD133, CD7, CD15, CD34, CD44, CD206, CD271, CEA (CarcinoEmbryonic Antigen), CGRP, chemokine receptors, cell-surface annexin-1, cell-surface plectin-1, Cripto-1, CRLR, CXCR2, CXCR4, DCC, DLL3, E2 glycoprotein, EGFR, EGFRvIII, EMR1, Endosialin, EP2, EP4, EpCAM, EphA2, ET receptors, Fibronectin, Fibronectin ED-B, FGFR, frizzled receptors, GAGE1, GAGE2, GAGE3, GAGE4, GAGE5, GAGE6, GLP-1 receptor, G-protein coupled receptors

of the Family A (Rhodopsin-like), G-protein coupled receptors of the Family B (Secretin receptor-like) like), G-protein coupled receptors of the Family C (Metabotropic Glutamate Receptor-like), GD2, GP100, GP120, Glypican-3, hemagglutinin, Heparin sulfates, HER1, HER2, HER3, HER4, HMFG, HPV 16/18 and E6/E7 antigens, hTERT, IL11-R, IL-13R, ITGAM, Kalikrien-9, Lewis Y, LH receptor, LHRH-R, LPA1, MAC-1, MAGE 1, MAGE 2, MAGE 3, MAGE 4, MART1, MC1R, Mesothelin, MUC1, MUC16, Neu (cell-surface Nucleolin), Neprilysin, Neuropilin-1, Neuropilin-2, NG2, NK1, NK2, NK3, NMB-R, Notch-1, NY-ESO-1, OT-R, mutant p53, p97 melanoma antigen, NTR2, NTR3, p32 (p32/gC1q-R/HABP1), p75, PAC1, PAR1, Patched (PTCH), PDGFR, PDFG receptors, PDT, Protease-cleaved collagen IV, proteinase 3, prohibitin, protein tyrosine kinase 7, PSA, PSMA, purinergic P2X family (*e.g.* P2X1-5), mutant Ras, RAMP1, RAMP2, RAMP3 patched, RET receptor, plexins, smoothened, sst1, sst2A, sst2B, sst3, sst4, sst5, substance P, TEMs, T-cell CD3 Receptor, TAG72, TGFBR1, TGFBR2, Tie-1, Tie-2, Trk-A, Trk-B, Trk-C, TR1, TRPA, TRPC, TRPV, TRPM, TRPML, TRPP (*e.g.* TRPV1-6, TRPA1, TRPC1-7, TRPM1-8, TRPP1-5, TRPML1-3), TSH receptor, VEGF receptors (VEGFR1 or Flt-1, VEGFR2 or FLK-1/KDR, and VEGF-3 or FLT-4), voltage-gated ion channels, VPAC1, VPAC2, Wilms tumor 1, Y1, Y2, Y4, or Y5.

[0105] In some examples, the protein expressed in the tumor is HER1/EGFR, HER2/ERBB2, CD20, CD25 (IL-2Rα receptor), CD33, CD52, CD133, CD206, CEA, CEACAM1, CEACAM3, CEACAM5, CEACAM6, cancer antigen 125 (CA125), alpha-fetoprotein (AFP), Lewis Y, TAG72, Caprin-1, mesothelin, PDGF receptor, PD-1, PD-L1, CTLA-4, IL-2 receptor, vascular endothelial growth factor (VEGF), CD30, EpCAM, EphA2, Glypican-3, gpA33, mucins, CAIX, PSMA, folate-binding protein, gangliosides (such as GD2, GD3, GM1 and GM2), VEGF receptor (VEGFR), VEGFR2, VEGF-A, integrin αVβ3, integrin α5β1, ERBB3, MET, IGF1R, EPHA3, TRAILR1, TRAILR2, RANKL, FAP, tenascin, AFP, BCR complex, CD3, CD18, CD44, CTLA-4, gp72, HLA-DR 10 β, HLA-DR antigen, IgE, MUC-1, nuC242, PEM antigen, metalloproteinases, Ephrin receptor, Ephrin ligands, HGF receptor, CXCR4, CXCR4, Bombesin receptor, SK-1antigen, Bcr-abl, RET, MET, TRKB, TIE2, ALK, ROS, EML4-ALK, ROS1, BRAFV600E, SRC, c-KIT, PDGFR, mTOR, TSC1, TSC2, BTK, KIT, BRCA, CDK 4/6, JAK1, JAK2, BRAF, FLT-3, MEK1, MEK2, or SMO.

[0106] In some examples, the conjugate is targeted to a protein expressed in the tumor. In some examples, the cell, hyperplasia or tumor is irradiated at a wavelength from or from about 600 nm to about 850 nm. In some examples, the tumor is irradiated at a wavelength of 690 ± 50 nm or 690 ± 20 nm.

[0107] In some examples, the cell, hyperplasia or tumor is irradiated at a dose of from or from about 2 J cm^{-2} to about 400 J cm^{-2} or from or from about 2 J/cm fiber length to about 500 J/cm fiber length. In some examples, the cells, hyperplasia or tumor are irradiated at a dose of at least or at least about 2 J cm^{-2}, 5 J cm^{-2}, 10 J cm^{-2}, 25 J cm^{-2}, 50 J cm^{-2}, 75 J cm^{-2}, 100 J cm^{-2}, 150 J cm^{-2}, 200 J cm^{-2}, 300 J cm^{-2}, 400 J cm^{-2}, or 500 J cm^{-2}; or the cells, tumor or hyperplasia are irradiated at a dose of at least or at least about 2 J/cm fiber length, 5 J/cm fiber length, 10 J/cm fiber length, 25 J/cm fiber length, 50 J/cm fiber length, 75 J/cm fiber length, 100 J/cm fiber length, 150 J/cm fiber length, 200 J/cm fiber length, 250 J/cm fiber length, 300 J/cm fiber length, 400 J/cm fiber length or 500 J/cm fiber length.

[0108] In some examples, the disease or condition is a tumor and the tumor is a superficial tumor. In some examples, the tumor is irradiated at a dose of at least or about at least or about10 J/cm^2, 25 J/cm^2, 50 J/cm^2, 150 J/cm^2, or 250 J/cm^2.

[0109] In some examples, the disease or condition is a tumor and the tumor is an interstitial tumor. In some examples, the tumor is irradiated at a dose of at least or about at least or about 50 J/cm fiber length, 100 J/cm fiber length, 200 J/cm fiber length, or 300 J/cm fiber length.

[0110] In some examples, the cells, hyperplasia or tumor is irradiated within or within about or about 12 hours, 24 hours, 36 hours, 72 hours or 96 hours after administering the conjugate. In some examples, the targeting molecule is administered up to 96 hours prior to administration of the conjugate. In some examples, the conjugate is administered in an amount that is from or from about 0.5 mg/kg to about 100 mg/kg or 20 mg/m^2 to about 4000 mg/m^2.

[0111] In some examples, the conjugate is administered in an amount that is at least or about at least or is or is about 0.5 mg/kg, 2.0 mg/kg, 4.0 mg/kg, 8.0 mg/kg, 16.0 mg/kg, 32.0 mg/kg or 64 mg/kg; or the conjugate is administered in an amount that is at least or about at least or is or is about 20 mg/m^2, 40 mg/m^2, 160 mg/m^2, 320 mg/m^2, 640 mg/m^2, 1280 mg/m^2 or 2560 mg/m^2.

[0112] In some examplesof the methods provided herein, prior to administration of the conjugate the targeting molecule is administered, *e.g.*, administered to the subject or the sample. In some examples, the targeting molecule is administered at a dose within a range from or from about 10 mg/m^2 to about 500 mg/m^2.

[0113] In some examples, the targeting molecule is an antibody or antigen binding fragment. In some examples, the antibody is cetuximab.

[0114] Provided in some examplesis a conjugate containing a phthalocyanine dye and a targeting molecule. In some such examples, the targeting molecule is a tissue-specific homing peptide, an RGD polypeptide, an iRGD polypeptide, a Lyp-1 polypeptide, a cripto-1 binding polypeptide, a somatostatin receptor binding polypeptide, a prohibitin binding polypeptide, a NGR polypeptide, an iNGR polypeptide, an activatable cell penetrating peptide (ACPP) comprised of a polycationic cell penetrating peptide (CPP) connected via a cleavable linker to a neutralizing polyanion, or an antibody, such as Ado-trastuzumab emtansine, Afatinib, Axitinib, Bosutinib, Cabozantinib, Ceritinib, Crizotinib, Dabrafenib, Dasatinib, Everolimus, Ibrutinib, Imatinib, Lenvatinib, Nilotinib, Olaparib, Palbociclib, Pazopanib, Ramucirumab, Regorafenib, Ruxolitinib, Sorafenib, Sunitinib, Temsirolimus, Trametinib, Vandetanib, Vemurafenib, Vismodegib, Ipilimumab, Nivolumab, pembrolizumab, MPDL3280A, Pidilizumab (CT-011), AMP-224, MSB001078C, or MEDI4736, or an antigen-binding fragment thereof.

[0114a] In yet another aspect, the present invention provides a conjugate, comprising a phthalocyanine dye and a targeting molecule, wherein the targeting molecule is selected from among a tissue-specific homing peptide, an RGD polypeptide, an iRGD polypeptide, a Lyp-1 polypeptide, a cripto-1 binding polypeptide, a somatostatin receptor binding polypeptide, a prohibitin binding polypeptide, a NGR polypeptide, an iNGR polypeptide, an activatable cell penetrating peptide (ACPP) comprised of a polycationic cell penetrating peptide (CPP) connected via a cleavable linker to a neutralizing polyanion, and an antibody selected from Ado-trastuzumab emtansine, Afatinib, Axitinib, Bosutinib, Cabozantinib, Ceritinib, Crizotinib, Dabrafenib, Dasatinib, Everolimus, Ibrutinib, Imatinib, Lenvatinib, Nilotinib, Olaparib, Palbociclib, Pazopanib, Ramucirumab, Regorafenib, Ruxolitinib, Sorafenib, Sunitinib, Temsirolimus, Trametinib, Vandetanib, Vemurafenib, Vismodegib, Ipilimumab, Nivolumab, pembrolizumab, pidilizumab, lambrolizumab, BMS-935559, MPDL3280A, Pidilizumab (CT-011), AMP-224, MSB001078C, or MEDI4736, or an antigen-binding fragment thereof.

[0115] In some examples, the ACPP comprises the structure: A-X1-B-, wherein B is a peptide portion of about 5 to about 20 basic amino acid residues, which is suitable for cellular uptake; A is a peptide portion of about 2 to about 20 acidic amino acid residues, which when linked with portion B is effective to inhibit or prevent cellular uptake of portion B; X1 is a cleavable linker of about 2 to about 100 atoms; and one or more of L-Y is linked to the C-terminus of peptide portion B.

[0116] In some examples, the homing peptide has a sequence as set forth in any of SEQ ID NOs: 1-52. In some examples, the dye is IR700.

Brief Description of the Drawings

[0117] **FIG.1A** displays the relative amounts of high molecular weight species (aggregates) and monomer form of the Cetuximab-IRDye 700DX as assessed by High Pressure Liquid Chromatography using a Size Exclusion column (HPLC-SEC) of a sample in a clear vial before 24 hours of light exposure.

CLAIMS

1. A method of manufacturing a phthalocyanine dye-targeting molecule conjugate, comprising:

a) contacting a targeting molecule with a phthalocyanine dye under conditions to produce a conjugate comprising the phthalocyanine dye linked to the targeting molecule; and

b) formulating the conjugate in a pharmaceutically acceptable buffer,

wherein in each of steps a) – b):

the only light to which the dye and conjugate are exposed has a wavelength within a range from about 400 nm to about 650 nm, or

the only light to which the dye and conjugate are exposed has an intensity of less than 500 lux.

2. The method of claim 1, wherein the contacting step a) further comprises contacting the targeting molecule with the phthalocyanine dye at a molar ratio of dye to targeting molecule of from or from about 1:1 to 100:1, and wherein the conjugate is formulated to a concentration from or from about 0.1 mg/mL to about 200.0 mg/mL.

3. The method of claim 1 or claim 2, wherein the conjugate is formulated to a concentration from or from about 0.5 mg/mL to about 10.0 mg/mL.

4. The method of any of claims 1-3, wherein the phthalocyanine dye comprises IRDye 700DX-NHS (IR700-NHS).

5. The method of any of claims 1-4, wherein the targeting molecule binds to an antigen, protein or cell surface target molecule.

6. The method of claim 5, wherein the cell surface target molecule is selected from among cell membrane phospholipids, prokaryotic peptidoglycans, bacterial cell envelop proteins, viral capsid proteins, ACTHR, endothelial cell Anxa-1, aminopetidase N, anti-IL-6R, alpha-4-integrin, alpha-5-beta-3 integrin, alpha-5-beta-5 integrin, alpha-fetoprotein (AFP), ANPA, ANPB, APA, APN, APP, 1AR, 2AR, AT1, B 1, B2, BAGE1, BAGE2, B-cell receptor BB 1, BB2, BB4, calcitonin receptor, cancer antigen 125 (CA 125), CCK1, CCK2,

CD5, CD10, CD11a, CD13, CD14, CD19, CD20, CD22, CD25, CD30, CD33, CD38, CD45, CD52, CD56, CD68, CD90, CD133, CD7, CD15, CD34, CD44, CD206, CD271, CEA (CarcinoEmbryonic Antigen), CGRP, chemokine receptors, cell-surface annexin-1, cell-surface plectin-1, Cripto-1, CRLR, CXCR2, CXCR4, DCC, DLL3, E2 glycoprotein, EGFR, EGFRvIII, EMR1, Endosialin, EP2, EP4, EpCAM, EphA2, ET receptors, Fibronectin, Fibronectin ED-B, FGFR, frizzled receptors, GAGE1, GAGE2, GAGE3, GAGE4, GAGE5, GAGE6, GLP-1 receptor, G-protein coupled receptors of the Family A (Rhodopsin-like), G-protein coupled receptors of the Family B (Secretin receptor-like) like), G-protein coupled receptors of the Family C (Metabotropic Glutamate Receptor-like), GD2, GP100, GP120, Glypican-3, hemagglutinin, Heparin sulfates, HER1, HER2, HER3, HER4, HMFG, HPV 16/18 and E6/E7 antigens, hTERT, IL11-R, IL-13R, IL-2 receptor, CTLA-4, PD-L1, ITGAM, Kalikrien-9, Lewis Y, LH receptor, LHRH-R, LPA1, MAC-1, MAGE 1, MAGE 2, MAGE 3, MAGE 4, MART1, MC 1R, Mesothelin, MUC 1, MUC 16, Neu (cell-surface Nucleolin), Neprilysin, Neuropilin-1, Neuropilin-2, NG2, NK1, NK2, NK3, NMB-R, Notch-1, NY-ESO-1, OT-R, mutant p53, p97 melanoma antigen, NTR2, NTR3, p32 (p32/gC1q-R/HABP1), p75, PAC1, PAR1, Patched (PTCH), PDGFR, PDFG receptors, PDT, Protease-cleaved collagen IV, proteinase 3, prohibitin, protein tyrosine kinase 7, PSA, PSMA, purinergic P2X family (*e.g.* P2X1-5), mutant Ras, RAMP1, RAMP2, RAMP3 patched, RET receptor, plexins, smoothened, sst1, sst2A, sst2B, sst3, sst4, sst5, substance P, TEMs, T-cell CD3 Receptor, TAG72, TGFBR1, TGFBR2, Tie-1, Tie-2, Trk-A, Trk-B, Trk-C, TR1, TRPA, TRPC, TRPV, TRPM, TRPML, TRPP (*e.g.* TRPV1-6, TRPA1, TRPC1-7, TRPM1-8, TRPP1-5, TRPML1-3), TSH receptor, VEGF receptors (VEGFR1 or Flt-1, VEGFR2 or FLK-1/KDR, and VEGF-3 or FLT-4), voltage-gated ion channels, VPAC1, VPAC2, Wilms tumor 1, Y1, Y2, Y4, and Y5.

7. The method of any of claims 1-6, wherein the targeting molecule is an antibody or an antibody fragment.

8. The method of any of claims 1-7, wherein the targeting molecule is cetuximab.

9. A stable conjugate, comprising the phthalocyanine dye-targeting molecule conjugate manufactured by the method of any one of claims 1-8, wherein the conjugate is stable for greater than three months or greater than 6 months or greater than 12 months.

10. The stable conjugate of claim 9, wherein the conjugate is stable if it retains greater than or greater than about 30%, 40%, 50%, 60%, 70%, 80%, 90% or more of its potency, activity or purity for greater than 3 months of storage compared to the conjugate's potency, activity or purity prior to storage.

11. A pharmaceutical composition comprising the stable conjugate of claim 9 or claim 10 and a pharmaceutically acceptable excipient.

12. The pharmaceutical composition of any of claims 9-11, wherein greater than 90% of the conjugate is present as a main monomer component.

13. A packaging system comprising the stable conjugate of claim 9 or claim 10, and further comprising a container, wherein the container protects from transmission of light such that the percentage of transmission of light is less than 50%.

14. The packaging system of claim 13, wherein the percentage of transmission of light is less than 10%.

15. The packaging system of claim 13 or claim 14, wherein the container protects from transmission of light having a wavelength from or from about 250 nm to about 800 nm.

16. Use of a phthalocyanine dye-targeting molecule conjugate manufactured according to the method of any one of claims 1-8 for the preparation of a medicament for use in a treatment regimen for treating a hyperplasia or a tumor in a subject, the treatment regimen comprising:

a) administering to the subject a therapeutically effective amount of the phthalocyanine dye-targeting molecule conjugate, wherein prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux; and

b) irradiating the hyperplasia or the tumor at a wavelength of 660 to 740 nm at a dose of at least 1 J cm or 1 J/cm of fiber length thereby treating the tumor in the subject.

17. Use of the stable conjugate of claim 9 or claim 10 for the preparation of a medicament for use in a treatment regimen for treating a hyperplasia or a tumor in a subject, the treatment regimen comprising:

a) administering to the subject a therapeutically effective amount of the stable conjugate, wherein prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux; and

b) irradiating the hyperplasia or the tumor at a wavelength of 660 to 740 nm at a dose of at least 1 J cm or 1 J/cm of fiber length thereby treating the tumor in the subject.

18. A pharmaceutical composition of claim 11 or claim 12 for use in a treatment regimen for treating a hyperplasia or a tumor in a subject, the treatment regimen comprising:

a) administering to the subject a therapeutically effective amount of the phthalocyanine dye-targeting molecule conjugate, wherein prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux; and

b) irradiating the hyperplasia or the tumor at a wavelength of 660 to 740 nm at a dose of at least 1 J cm or 1 J/cm of fiber length thereby treating the tumor in the subject.

19. The pharmaceutical composition of claim 11 or claim 12 for use in a treatment regimen for treating a hyperplasia or a tumor in a subject, the treatment regimen comprising:

a) administering to the subject a therapeutically effective amount of the stable conjugate, wherein prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux; and

b) irradiating the hyperplasia or the tumor at a wavelength of 660 to 740 nm at a dose of at least 1 J cm or 1 J/cm of fiber length thereby treating the tumor in the subject.

20. A method of removing unwanted cells or pathogens in a sample comprising:

a) administering to a sample, the sample comprising unwanted cells or pathogens, a therapeutically effective amount of the conjugate manufactured according to the method of any of claims 1-8, wherein prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux; and

b) irradiating the unwanted cells or pathogens at a wavelength of 660 to 740 nm at a dose of at least 1 J cm or 1 J/cm of fiber length thereby removing the unwanted cells or pathogens in the sample, optionally wherein the method is performed *in vitro* or *ex vivo*.

21. Use of a phthalocyanine dye-targeting molecule conjugate manufactured according to the method of any one of claims 1-8 for the preparation of a medicament for use in a treatment regimen for removing unwanted cells or pathogens in a subject, wherein the unwanted cells or pathogens are associated with a disease or condition in the subject and the treatment regimen comprising:

a) administering to the subject a therapeutically effective amount of the conjugate, wherein prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux; and

b) irradiating the unwanted cells or pathogens at a wavelength of 660 to 740 nm at a dose of at least 1 J cm or 1 J/cm of fiber length thereby removing the unwanted cells or pathogens in the subject and treating the disease or condition in the subject.

22. A pharmaceutical composition of claim 11 or claim 12 for use in a treatment regimen for removing unwanted cells or pathogens in a subject, wherein the unwanted cells or pathogens are associated with a disease or condition in the subject and the treatment regimen comprising:

a) administering to the subject a therapeutically effective amount of the conjugate, wherein prior to and during the administration step the conjugate is not exposed to an intensity of environmental light greater than 500 lux; and

b) irradiating the unwanted cells or pathogens at a wavelength of 660 to 740 nm at a dose of at least 1 J cm or 1 J/cm of fiber length thereby removing the unwanted cells or pathogens in the subject and treating the disease or condition in the subject.

23. The use of claim 21 or pharmaceutical composition for use of claim 22, wherein disease or condition is a hyperplasia or tumor, an infection, an inflammatory disease or condition, or a neuronal disease or condition.

24. A conjugate, comprising a phthalocyanine dye and a targeting molecule, wherein the targeting molecule is selected from among a tissue-specific homing peptide, an RGD polypeptide, an iRGD polypeptide, a Lyp-1 polypeptide, a cripto-1 binding polypeptide, a somatostatin receptor binding polypeptide, a prohibitin binding polypeptide, a NGR polypeptide, an iNGR polypeptide, an activatable cell penetrating peptide (ACPP)

comprised of a polycationic cell penetrating peptide (CPP) connected via a cleavable linker to a neutralizing polyanion, and an antibody selected from Ado-trastuzumab emtansine, Afatinib, Axitinib, Bosutinib, Cabozantinib, Ceritinib, Crizotinib, Dabrafenib, Dasatinib, Everolimus, Ibrutinib, Imatinib, Lenvatinib, Nilotinib, Olaparib, Palbociclib, Pazopanib, Ramucirumab, Regorafenib, Ruxolitinib, Sorafenib, Sunitinib, Temsirolimus, Trametinib, Vandetanib, Vemurafenib, Vismodegib, Ipilimumab, Nivolumab, pembrolizumab, pidilizumab, lambrolizumab, BMS-935559, MPDL3280A, Pidilizumab (CT-011), AMP-224, MSB001078C, or MEDI4736, or an antigen-binding fragment thereof.