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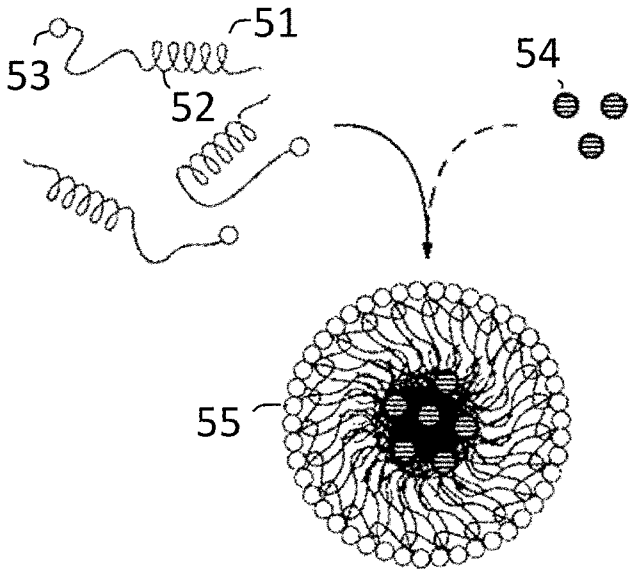

FIG. 1

(57) Abstract: In a first aspect, the present invention relates to the use of a micelle of an amphiphilic block copolymer to modify a cell, wherein the amphiphilic block copolymer comprises one or more hydrophobic blocks and one or more hydrophilic blocks, wherein at least one block comprises a conjugated polymer, and wherein the micelle is optionally loaded with a compound therein.

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BLOCK COPOLYMER MICELLES

Technical field of the invention

The present invention relates to the field of cell modification and in particular to the use of micelles formed from amphiphilic block copolymers to modify cells.

Background of the invention

Nanoparticles, such as micelles, liposomes or vesicles, comprising polymers have been under considerable investigation for various uses in biomedical applications. For example, efforts have been made towards their use as imaging probes, compound carriers and for tissue or cell specific targeting. Furthermore, nanoparticles fulfilling a plurality of these functions are desirable. Polymers are an interesting class of materials in these applications, as multiple biocompatible polymers are known and as they can be made to fulfil a multitude of different functions.

Amphiphilic block copolymers have for example been used as surfactants to form micelles. These can be loaded with e.g. dyes or drugs, which can in turn be released when specific triggers are fulfilled. Furthermore, the amphiphilic block copolymers or micelle surfaces themselves may be further functionalized with dyes or specific targeting groups. However, particularly when multiple functions are to be combined, these applications typically have in common that they are fairly complex: for example block copolymers comprising three or more blocks, additional functionalisation with e.g. a dye, crosslinking to stabilize the micelles and their content, release triggers which are difficult to achieve, etc. may all be required.

Alternatively, conjugated polymers have also been used as dyes loaded in micelles based on other surfactants. These kinds of micelles are for example described in US 2012/0269736 A1. However, this approach already requires two different materials to obtain a fairly simple imaging probe and does not lend itself well to incorporating additional functionalities.

There is thus still a need for simple micellar systems based on polymers, which may be cheap to produce and easy to process, for use as imaging probes in biomedical applications and which may additional easily double up as drug or other compound delivery systems.

Summary of the invention

It is an object of the present invention to provide good amphiphilic block copolymer based micelles for use to modify a cell.

It is an advantage of embodiments of the present invention that micelles can be provided that can be used as imaging probes in biomedical applications.

It is an advantage of embodiments of the present invention that the block copolymer is an amphiphilic block copolymer that may be used as the surfactant that forms the micelles.

It is an advantage of embodiments of the present invention that the copolymer itself may be used as a dye or other contrast agent. In particular, it is an advantage of embodiments of the present invention that micelles made of amphiphilic block copolymers comprising one or more hydrophobic conjugated polymer blocks and one or more hydrophilic blocks, wherein at least one block comprises a conjugated

polymer, display no fluorescence in water as long as they are outside of a cell, but rapidly start to fluoresce once inside a cell. This surprising property which, to the best of our knowledge, has been observed by micelles for the first time, is particularly useful as it gives an independent signature that a micelle has entered a cell and that the micelle has released its optional content, even if that content is itself not fluorescent or otherwise marked. Without being bound by theory, the inventors believe that one possible explanation for this phenomena is that such block copolymers do not fluoresce in their micellar phase due to inter-molecular π–π stacking of the individual block copolymers forming the micelles. Still not being bound by theory, the inventors believe that the fluorescence appearing rapidly after the micelles enter the cells is an indication that the π–π stacking is broken during entry in the cell and/or shortly thereafter. It is a further advantage of embodiments of the present invention that the appearance of fluorescence after cellular uptake allows one to gain information about the fate of micelles over a period of time.

It is an advantage of embodiments of the present invention that the amphiphilic block polymers may be relatively simple and cheap to produce.

It is an advantage of embodiments of the present invention that the micelles may be easy and cheap to process.

It is an advantage of embodiments of the present invention that the micelles may be stable in an aqueous colloid for an extended period of time.

It is an advantage of embodiments of the present invention that the micelles are easily internalized into (e.g. living) cells.

It is an advantage of embodiments of the present invention that the micelles may be loaded with a compound.

It is an advantage of embodiments of the present invention that the loaded compound may be readily released inside the cell. Furthermore, no particular external trigger needs to be applied for this release to occur.

The above objective is accomplished by a method and device according to the present invention.

In a first aspect, the present invention relates to the use of a micelle of an amphiphilic block copolymer to modify a cell, wherein the amphiphilic block copolymer comprises one or more hydrophobic blocks and one or more hydrophilic blocks, wherein at least one block comprises a conjugated polymer, and wherein the micelle is optionally loaded with a compound therein.

In a second aspect, the present invention relates to a micelle of an amphiphilic block copolymer usable according to the first aspect, the amphiphilic block copolymer comprising:

i. a first block comprising a poly(*p*-arylene vinylene) and
ii. a non-ionic second block comprising a vinyl polymer;

wherein at least one block is hydrophobic and at least one other block is hydrophilic.

In a third aspect, the present invention relates to a method for forming the micelle of the second aspect, comprising the steps of:

a. dissolving the amphiphilic block copolymer in a first liquid and
b. mixing thereto a second liquid;

wherein either the first or second liquid is an aqueous liquid and wherein the other liquid is a non-aqueous liquid.

In a fourth aspect, the present invention relates to an amphiphilic block copolymer usable to make the micelle of the third aspect, comprising:

i. a first block comprising a poly(*p*-arylene vinylene) and
ii. a nonionic second block comprising a vinyl polymer;

wherein at least one block is hydrophobic and at least one other block is hydrophilic.

In a fifth aspect, the present invention relates to a method for synthesizing an amphiphilic block copolymer in accordance with the fourth aspect, comprising the steps of:

a. providing an initiator comprising a first initiating moiety for an anionic polymerization and a second initiating moiety for a living radical polymerization,
b. polymerizing, in an anionic initiation pathway, a poly(*p*-arylene vinylene) monomer with the first initiating moiety, so as to form a precursor to a poly(*p*-arylene vinylene) first block,
c. polymerizing, in a living radical polymerization pathway, a vinyl monomer with the second initiating moiety, so as to form a second block, and
d. performing a thermal elimination, so as to obtain the poly(*p*-arylene vinylene) first block.

Particular and preferred aspects of the invention are set out in the accompanying independent and dependent claims. Features from the dependent claims may be combined with features of the independent claims and with features of other dependent claims as appropriate and not merely as explicitly set out in the claims.

Although there has been constant improvement, change and evolution of devices in this field, the present concepts are believed to represent substantial new and novel improvements, including departures from prior practices, resulting in the provision of more efficient, stable and reliable devices of this nature.

The above and other characteristics, features and advantages of the present invention will become apparent from the following detailed description, taken in conjunction with the accompanying drawings, which illustrate, by way of example, the principles of the invention. This description is given for the sake of example only, without limiting the scope of the invention. The reference figures quoted below refer to the attached drawings.

Brief description of the drawings

Fig. 1 shows a schematic representation of an embodiment of the present invention wherein an amphiphilic block copolymer and, optionally, an additional compound form a (loaded) micelle.

Fig. 2 to 11 show synthetic reaction schemes to obtain amphiphilic block copolymers according to embodiments of the present invention.

Fig. 12 shows the cytotoxicity of unloaded micelles according to embodiments of the present invention.

Fig. 13 shows the cytotoxicity of loaded micelles according to embodiments of the present invention.

In the different figures, the same reference signs refer to the same or analogous elements.

Description of illustrative embodiments

The present invention will be described with respect to particular embodiments and with reference to certain drawings but the invention is not limited thereto but only by the claims. The drawings described are only schematic and are non-limiting. In the drawings, the size of some of the elements may be exaggerated and not drawn on scale for illustrative purposes. The dimensions and the relative dimensions do not correspond to actual reductions to practice of the invention.

Furthermore, the terms first, second, third and the like in the description and in the claims, are used for distinguishing between similar elements and not necessarily for describing a sequence, either temporally, spatially, in ranking or in any other manner. It is to be understood that the terms so used are interchangeable under appropriate circumstances and that the embodiments of the invention described herein are capable of operation in other sequences than described or illustrated herein.

Moreover, the terms top, bottom, over, under and the like in the description and the claims are used for descriptive purposes and not necessarily for describing relative positions. It is to be understood that the terms so used are interchangeable under appropriate circumstances and that the embodiments of the invention described herein are capable of operation in other orientations than described or illustrated herein.

It is to be noticed that the term "comprising", used in the claims, should not be interpreted as being restricted to the means listed thereafter; it does not exclude other elements or steps. It is thus to be interpreted as specifying the presence of the stated features, integers, steps or components as referred to, but does not preclude the presence or addition of one or more other features, integers, steps or components, or groups thereof. Thus, the scope of the expression "a device comprising means A and B" should not be limited to devices consisting only of components A and B. It means that with respect to the present invention, the only relevant components of the device are A and B.

Reference throughout this specification to "one embodiment" or "an embodiment" means that a particular feature, structure or characteristic described in connection with the embodiment is included in at least one embodiment of the present invention. Thus, appearances of the phrases "in one embodiment" or "in an embodiment" in various places throughout this specification are not necessarily all referring to the same embodiment, but may. Furthermore, the particular features, structures or characteristics may be combined in any suitable manner, as would be apparent to one of ordinary skill in the art from this disclosure, in one or more embodiments.

Similarly it should be appreciated that in the description of exemplary embodiments of the invention, various features of the invention are sometimes grouped together in a single embodiment, figure, or description thereof for the purpose of streamlining the disclosure and aiding in the understanding of one or more of the various inventive aspects. This method of disclosure, however, is not to be interpreted as reflecting an intention that the claimed invention requires more features than are expressly recited in each claim. Rather, as the following claims reflect, inventive aspects lie in less than all features

of a single foregoing disclosed embodiment. Thus, the claims following the detailed description are hereby expressly incorporated into this detailed description, with each claim standing on its own as a separate embodiment of this invention.

Furthermore, while some embodiments described herein include some but not other features included in other embodiments, combinations of features of different embodiments are meant to be within the scope of the invention, and form different embodiments, as would be understood by those in the art. For example, in the following claims, any of the claimed embodiments can be used in any combination.

In the description provided herein, numerous specific details are set forth. However, it is understood that embodiments of the invention may be practiced without these specific details. In other instances, well-known methods, structures and techniques have not been shown in detail in order not to obscure an understanding of this description.

As used herein and unless provided otherwise, a micelle of an amphiphilic block copolymer is a micelle formed of amphiphilic block copolymers.

As used herein and unless provided otherwise, a block copolymer suitable for any aspect of the present invention comprises at least one hydrophobic block and at least one hydrophilic block. If the block copolymer comprises more than one hydrophobic block, they are preferably directly linked to each other. If the block copolymer comprises more than one hydrophilic block, they are preferably at the extremities of the block copolymer. In an embodiment, the block copolymer can be a triblock comprising a hydrophobic block connected to two hydrophilic blocks. Preferably, the block copolymer is a diblock comprising a single hydrophilic block linked to a single hydrophobic block.

As used herein and unless provided otherwise, a hydrophobic block of a block copolymer is a polymeric block of such a chemical nature and length that a homopolymer of this chemical nature and length would not be soluble in water.

As used herein and unless provided otherwise, a hydrophilic block of a block copolymer is a polymeric block of such a chemical nature and length that a homopolymer of this chemical nature and length would be soluble at a concentration of at least 1 wt% in water.

As used herein with respect to a linking group, and unless otherwise stated, the term 'arylene' designates any divalent group derived from aryl (such as below defined) by abstracting a hydrogen atom.

As used herein with respect to a linking group, and unless otherwise stated, the term 'heteroarylene' designates any divalent group derived from heteroaryl (such as below defined) by abstracting a hydrogen atom.

As used herein with respect to a substituting group, and unless otherwise stated, the terms 'homocyclic aromatic' or 'aryl' designate any mono- or polycyclic aromatic monovalent hydrocarbon group having from 6 to 15 carbon atoms such as, but not limited to, phenyl, naphthyl, anthracenyl, phenanthracyl, fluoranthenyl, chrysenyl, pyrenyl, biphenylyl, terphenyl, picenyl, indenyl, biphenyl, indacenyl, tetrahydropyrenyl, benzocyclobutenyl, benzocyclooctenyl and the like, including fused benzo-C_{4-8} cycloalkyl groups such as, for instance, indanyl, tetrahydronaphthyl, fluorenyl and the like, all of the said groups being optionally substituted with one or more substituents (preferably 1 to 3 substituents) independently selected from the group consisting of halogen, C_{1-12} alkyl, nitro, trifluoromethoxy,

trifluoromethyl and C_{1-12} alkoxy (all of such substituents being such as herein defined, including individual species and sub-groups thereof), such as, but not limited to, 4-fluorophenyl, 4-chlorophenyl, 3,4-dichlorophenyl, 2,6-dichlorophenyl, 2-fluorophenyl, 3-chlorophenyl, 3,5-dichlorophenyl, trifluoromethylphenyl, 3,4-dimethoxyphenyl, iodophenyl, and bromophenyl.

As used herein with respect to a substituting group, and unless otherwise stated, the terms 'heterocyclic aromatic' or 'heteroaryl' means a mono- or polycyclic, polyunsaturated, monovalent hydrocarbon group having from 4 to 12 carbon atoms and including one or more heteroatoms in one or more heterocyclic rings, each of said rings having 5 or 6 atoms (and optionally further including one or more heteroatoms attached to one or more carbon atoms of said ring, for instance in the form of a carbonyl, and/or to one or more heteroatoms of said ring, for instance in the form of a N-oxide), each of said heteroatoms being independently nitrogen or sulfur, also including groups wherein a heterocyclic ring is fused to one or more aromatic homocyclic rings for instance in the form of benzo-fused, dibenzo-fused and naphtho-fused heterocyclic groups; within this definition are included heteroaryl groups such as, but not limited to, thienyl, pyrrolyl, pyridyl, carbazolyl and benzothiazolyl.

The arylene or heteroarylene divalent group Ar may be selected from the group consisting of 1,4-phenylene; 2,6-naphthalenediyl; 1,4-naphthalenediyl; 1,4-anthracenediyl; 2,6-anthracenediyl; 9,10-anthracenediyl; 2,5-thienylene; 2,5-furanediyl; 2,5-pyrrolediyl; 1,3,4-oxadiazole-2,5-dyil; 1,3,4-thiadiazole-2,5-diyl; 2,3-benzo[c]thienylene; thieno[3,2-b]thiophene-2,5-diyl; pyrrolo[3,2-b]pyrrole-2,5-diyl; pyrene-2,7-diyl; 4,5,9,10-tetrahydropyrene-2,7-diyl; 4,4'-bi-phenylene; phenantrene-2,7-diyl; 9,10-dihydrophenantrene-2,7-diyl; dibenzo-furane-2,7-diyl; dibenzothiophene-2,7-diyl; 2,5-selenophenylene; isothianaphthylene; 2,7-silafluorenylene; 3,6-carbazolediyl; pyridinediyl; 2,2'-dipyridinediyl; pyridopyrazinediyl; quinoxalinediyl; thieno[3,4-c]pyridinediyl; thieno[3,4-b]pyridinediyl; thieno[3,4-b]pyrazinediyl; benzothiadiazolediyl; 4H-cyclopenta[2,1-b;3,4-b']dithienylene; silacyclopentadienediyl; and anthrazolinediyl.

As used herein with respect to a substituting group, and unless otherwise stated, the term 'C_{1-12} alkyl' refers to a straight (non-branched) or branched chain saturated acyclic hydrocarbon monovalent group having from 1 to 4 carbon atoms such as, for example, methyl, ethyl, propyl, n-butyl, 1-methylethyl (isopropyl), 2-methylpropyl (isobutyl), and 1,1-dimethylethyl (*tert*-butyl). Similarly, the term '' C_{1-20} alkyl '' refers to straight (non-branched) or branched chain groups having from 1 to 20 carbon atoms such as, for example, pentyl, hexyl, heptyl, octyl, nonyl, decyl, dodecyl, tetradecyl, hexadecyl, octadecyl and the like.

As used herein with respect to a substituting group, and unless otherwise stated, the term "C_{1-12} alkoxy" refers to substituents wherein a carbon atom of a C_{1-20} alkyl group (such as defined herein above, including sub-groups thereof), is attached to an oxygen atom through a single bond, including methoxy, ethoxy, propoxy, n-butoxy, isopropoxy, sec-butoxy, and tert-butoxy.

In a first aspect, the present invention relates to the use of a micelle of an amphiphilic block copolymer to modify a cell, wherein the amphiphilic block copolymer comprises one or more hydrophobic

blocks and one or more hydrophilic blocks, wherein at least one block comprises a conjugated polymer, and wherein the micelle is optionally loaded with a compound therein.

We now refer to Fig. 1. An amphiphilic block copolymer (51) comprising at least a hydrophobic (52) and a hydrophilic (53) block can advantageously be used as a surfactant to form a micelle (54). Furthermore, at least one block comprises a conjugated polymer, i.e. a hydrophobic block or a hydrophilic block may comprise a conjugated polymer. Preferably, only one block comprises a conjugated polymer. More preferably, the block copolymer is a di- or a tri-block comprising a single hydrophobic block, said hydrophobic block comprising a conjugated polymer. Conjugated polymers are typically luminescent, such as photoluminescent, which advantageously allows them to act as contrast agents, such as dyes, for a variety of microscopic and spectroscopic techniques.

The cell may typically be a living cell, but it may equally be a dead cell, an artificial cell, such as an artificial lysosome, or even an extracellular vesicle.

The micelle may optionally be loaded with a compound (55), i.e. it may contain a compound. For example, an amphiphilic block copolymer may organize itself so as to form a hydrophilic shell around a hydrophobic core and the compound may be present inside the hydrophobic core. The compound referred to here is a compound which is different from the block copolymer forming the micelle. This compound may for example be a dye or another contrast agent, such as a contrast agent for magnetic resonance imaging, or it may be a drug or polynucleotide. In some embodiments, the compound may have a same hydrophobicity type as the core; i.e. both the core and the compound may be hydrophobic or both may be hydrophilic. In some embodiments wherein the micelles are dispersed in a liquid medium, the compound may have a water solubility of 5% or less, preferably 1% or less, yet more preferably 0.1% or less.

In embodiments, the conjugated polymer may be a poly(p-arylene vinylene), such as a poly(p-phenylene vinylene), or a poly(p-arylene ethynylene), poly(p-heteroarylene vinylene), poly(p-heteroarylene ethynylene), polythiophene, polyphenylene, polyfluorene, polyacetylene or polypyrole. Depending on the nature of the sidechains substituted thereon, a hydrophilic or a hydrophobic block may typical be obtained; regardless of the class of conjugated polymer which is selected.

In embodiments, the block copolymer may typically also comprise a non-conjugated polymer block, such as a poly(meth)acrylate, poly(meth)acrylamide, polystyrene, polyoxazoline, polyphosphate, polyvinylalcohol, polyacrylic acid, polyamines, polyvinylpyrrolidone, polyvinyl methyl ether-maleic anhydride or polyethyleneimine. Each of these classes of conjugated polymers may typically be used to form either a hydrophilic or a hydrophobic block, depending on the nature of the sidechains substituted thereon. Depending on the nature of the sidechains substituted thereon, a hydrophilic or a hydrophobic block may typical be obtained; regardless of the class of non-conjugated polymer which is selected

In embodiments, modifying the cell may comprise dyeing at least part of the cell and/or may comprise releasing the compound, if present, inside the cell.

Dyeing the cell, i.e. pigmenting at least a part of the cell with a luminescent or colored substance such as a luminescent polymer or compound, advantageously allows it to be better visualized through a variety of microscopic and spectroscopic techniques. Dyeing the cell may be achieved by means of the

amphiphilic block copolymer itself, e.g. through the conjugated polymer it comprises, and/or by means of the compound, if present, functioning as a dye or other contrast agent. The compound may however also have a different function, in which case releasing the compound advantageously allows the compound to fulfil this function.

In embodiments, the conjugated polymer may display a relatively low photoluminescence when the amphiphilic block copolymer is present in a micelle, but may display a higher photoluminescence when present in a more unaggregated form, e.g. after the micelle has broken up. Without being bound by theory, it is believed that the close stacking of the conjugated polymer chromophores when present in the micelle leads to photoluminescence quenching, e.g. due to exciton-exciton annihilation. Advantageously, photoluminescence of the conjugated polymer is typically observed when it is present in a cell. This suggests that a breaking up of the micelle typically occurs upon or after uptake in the cell, thereby advantageously also releasing the compound, if present, inside the cell.

In embodiments, the present invention may relate to the use of a micelle exhibiting a first photoluminescence intensity when illuminated at a wavelength, the micelle being formed of an amphiphilic block copolymer, to modify a cell by transferring in the cell the amphiphilic block copolymer in a non-micellar form, said block copolymer in a non-micellar form exhibiting a second photoluminescence intensity when illuminated at the wavelength, the second intensity being higher than the first intensity. The wavelength is selected in the absorption spectrum of the amphiphilic block copolymer, and preferably at the maximum of the UV-Vis absorption spectrum. In embodiments, the second photoluminescence may be at least ten times more intensive than the first photoluminescence. Preferably, the first photoluminescence is null.

In preferred embodiments, the block copolymer may be non-ionic. Ionic block copolymers typically lead to micelles which have a large tendency to aggregate in aqueous medium. Conversely, non-ionic block copolymers advantageously lead to micelles which do not display this large tendency.

In the first aspect, the micelle may be according to any embodiment of the second aspect.

In the first aspect, the amphiphilic block copolymer may be according to any embodiment of the fourth aspect.

In a second aspect, the present invention relates to a micelle of an amphiphilic block copolymer usable according to the first aspect, the amphiphilic block copolymer comprising:

i. a first block comprising a poly(*p*-arylene vinylene) and
ii. a second block comprising a vinyl polymer;

wherein at least one block is hydrophobic and at least one other block is hydrophilic.

A micelle of an amphiphilic block copolymer can advantageously be made (cf. infra) from an amphiphilic block copolymer, i.e. a block copolymer comprising at least a hydrophobic and a hydrophilic block, comprising a poly(*p*-arylene vinylene) in a first block, e.g. a hydrophobic block may comprise a poly(*p*-arylene vinylene) with hydrophobic side chains, and a vinyl polymer in a second block, e.g. a hydrophilic block may comprise a vinyl polymer with hydrophilic side chains.

In embodiments, the micelle may be present in an aqueous medium. In preferred embodiments, the micelle may be present in an aqueous medium comprising at least 90% water. For example, the micelle may be present in water.

In preferred embodiments, the micelle may be sufficiently stable in the aqueous medium for 6 hours or more, more preferably 24 hours or more, most preferably 1 month or more. The micelles may for example be sufficiently stable in the aqueous medium for more than 3 months, such as for more than one year or for more than 1.5 year.

As cells are typically present in an aqueous medium themselves, micelles present and stable in an aqueous medium are advantageously suitable for uptake by such a cell.

In embodiments, the poly(*p*-arylene vinylene) may be selected in such a way that it is luminescent and preferably photoluminescent in solution when in a non-micellar form.

A block copolymer comprising a luminescent, such as photoluminescent, poly(*p*-arylene vinylene) can advantageously by itself, i.e. without any additional loaded or functionalized compounds, act as a dye for a variety of microscopic or spectroscopic techniques.

In embodiments, the micelle may be loaded with a compound therein. In preferred embodiments, the micelle may be loaded with a loading efficiency of 5 % or more, preferably 10 % or more, yet more preferably 20 % or more, most preferably 30 % or more, such as up to 60 %. The loading efficiency as used herein may be defined as the ratio of the mass of compound loaded into the micelles to the total mass of compound initially added to the mixture.

The micelle can advantageously be loaded with a compound which can then in turn be released within a cell. A high loading advantageously allows more of the compound to be released within the cell.

In embodiments, the compound may be a contrast agent, a drug or a polynucleotide.

Contrast agents, such as dyes, advantageously allow an improved performance of a variety of microscopic and spectroscopic techniques. Drugs or polynucleotides advantageously allow other functions to be performed on or in a cell.

In preferred embodiments, the block copolymer may be non-ionic. Ionic block copolymers typically lead to micelles which have a large tendency to aggregate in aqueous medium. Conversely, non-ionic block copolymers advantageously lead to micelles which do not display this large tendency.

In the second aspect, the amphiphilic block copolymer may be according to any embodiment of the fourth aspect.

In the second aspect, the hydrophilic and/or hydrophobic block may be according to any embodiment of the fourth aspect.

In a third aspect, the present invention relates to a method for forming the micelle of the second aspect, comprising the steps of:

a. dissolving the amphiphilic block copolymer in a first liquid and
b. mixing thereto a second liquid;

wherein either the first or second liquid is an aqueous liquid and wherein the other liquid is a non-aqueous liquid.

Mixing a second liquid to a solution of the amphiphilic block copolymer in a first liquid, wherein one of the liquids is an aqueous liquid and the other liquid is a non-aqueous liquid, advantageously prompts the amphiphilic block copolymer to self-assemble into a micelle. When the second liquid is an aqueous medium, a micelle is typically obtained wherein the outer shell is hydrophilic and the core is hydrophobic. Conversely, when the second liquid is a non-aqueous liquid, inverse micelles comprising a hydrophobic outer shell and a hydrophilic core are typically obtained. The non-aqueous liquid may typically be a polar solvent, such as a polar aprotic sovent, e.g. DMF.

Mixing the second liquid to the first liquid may for example comprise slowly adding, such as dripping, the second liquid to the first liquid. In some embodiments, the speed at which the second liquid is added may influence the size of the obtained micelles. Adding the second liquid at a slower rate may for example lead to smaller micelles.

In embodiments, the first liquid may be a non-aqueous polar solvent, miscible with water and the second liquid may be water. Preferably, the first liquid may be non-aqueous polar aprotic solvent, miscible with water and the second liquid may be water. For instance, the first liquid may be dimethylformamide (DMF) and the second liquid may be water.

In embodiments, the method may comprise an additional step of:

c. removing the first liquid.

Removing the first liquid advantageously allows a colloid of the micelles in a pure continuous phase of the second liquid to be obtained, e.g. a colloid of micelles in water.

In embodiments, an additional compound may be present in the first and/or the second liquid, thereby forming the micelle loaded with the compound therein. In preferred embodiments, the compound may be present in the first liquid. In preferred embodiments, the compound may be present in the first and/or the second liquid before step b. In other embodiments, the compound may only be added to the first and the second liquid after step b. In embodiments, the compound may have a solubility of less than 1g/l into the second liquid.

The presence of the compound in the mixture advantageously allows it to be incorporated or loaded into the micelles. Having the compound present before the formation of the micelles in step b, typically advantageously facilitates the loading of the compound in the micelles. However, in other cases, a sufficiently high loading may be obtained by only adding the compound after the formation of the micelles in step b. Having a compound that has a low solubility into the second liquid, typically advantageously facilitates the loading of this compound into the micelles.

In a fourth aspect, the present invention relates to an amphiphilic block copolymer usable to make the micelle of the third aspect, comprising:

i. a first block comprising a poly(*p*-arylene vinylene) and

ii. a non-ionic second block comprising a vinyl polymer;

wherein at least one block is hydrophobic and at least one other block is hydrophilic.

In embodiments, the poly(*p*-arylene vinylene) may for example be a poly(*p*-phenylene vinylene).

Poly(*p*-arylene vinylenes), such as a poly(*p*-phenylene vinylene) (PPV), are conjugated polymers which are advantageously photoluminescent, non-toxic and whose hydrophobicity or hydrophilicity may be tuned through its side chains. Vinyl polymers, such as poly(meth)acrylate esters, poly(meth)acrylamides or polystyrenes are advantageously hydrophilic or hydrophobic polymers which can be non-toxic and which can be synthesized through a reaction scheme that is compatible with that of the poly(*p*-arylene vinylenes) (cf. infra). An amphiphilic block copolymer consisting of only non-toxic blocks may typically be advantageously non-toxic itself.

In embodiments, the block copolymer may be a di- or tri-block copolymer.

A di- or tri-block copolymer is advantageously a fairly simple compound which may be relatively easy and cheap to produce.

In embodiments, the poly(*p*-arylene vinylene) may comprise hydrophobic substituents.

In embodiments, the vinyl polymer may be obtainable by the polymerization of a monomer obtainable from the reaction of (meth)acrylic acid with respectively an alcohol of general formula HO-R or an amine of general formula NH_2-R, wherein R is non-ionic. R may for example be a C_{1-20} alkyl or alkoxy, such as a C_{1-12} alkyl or alkoxy, or a functionalized derivative thereof.

In embodiments, the vinyl polymer may be a hydrophilic poly(ethylene glycol monomethyl ether methacrylate) (PEGMA), a poly(2-hydroxyethyl acrylate) (PHEA), or a poly(n-(2-hydroxypropyl) methacrylamide) (HPMA), or a polystyrene (PS).

Particularly PEGMA, PHEA and PS are non-toxic vinyl polymers.

In embodiments, the poly(*p*-arylene vinylene) may be a poly(2-methoxy-5-(3,7-dimethyloctyloxy)-1,4-phenylene vinylene) or a poly(2-methoxy-5-(2-ethylhexyloxy)-1,4-phenylenevinylene).

In embodiments, the conjugated polymer may have a length of 7 repeating units or more. The conjugated polymer may for example have a length of up to 100, preferably up to 50, most preferably up to 15 repeating units.

In embodiments, the copolymer may be a poly(*p*-arylene vinylene)-*block*-poly(meth)acrylate ester, a poly(*p*-arylene vinylene)-*block*-poly(meth)acrylamide or a poly(*p*-arylene vinylene)-*block*-polystyrene.

Poly(*p*-arylene vinylene)-*block*-poly(meth)acrylate esters, poly(*p*-arylene vinylene)-*block*-poly(meth)acrylamides and poly(*p*-arylene vinylene)-*block*-polystyrenes are advantageously fairly simple amphiphilic di-block copolymers which can be non-toxic and which can be produced relatively easily and cheaply.

In a fifth aspect, the present invention relates to a method for synthesizing an amphiphilic block copolymer in accordance with the fourth aspect, comprising the steps of:

a. providing an initiator comprising a first initiating moiety for an anionic polymerization and a second initiating moiety for a living radical polymerization,

b. polymerizing, in an anionic initiation pathway, a poly(*p*-arylene vinylene)-monomer with the first initiating moiety, so as to form a precursor to a poly(*p*-arylene vinylene) first block,

c. polymerizing, in a living radical polymerization pathway, a vinyl monomer with the second initiating moiety, so as to form a second block, and

d. performing a thermal elimination, so as to obtain the poly(*p*-arylene vinylene) first block.

Using an initiator using two different initiating moieties advantageously allows a block copolymer, such a di-block copolymer, to be synthesized using two distinct polymerization pathways: on the one hand a precursor to a first block, such as a PPV precursor, through an anionic initiation pathway and on the other hand a second block through a living radical polymerization, such as a single electron transfer living radical polymerization. The first block precursor, such as the PPV-precursor, can subsequently be transformed into the final first block, such as the final PPV block, through a thermal elimination step. Depending on the embodiment, this thermal elimination may be performed before or after the living radical polymerization.

In embodiments, the poly(*p*-arylene vinylene) may be a poly(*p*-phenylene vinylene).

In embodiments, the initiator may be 4-((methylsulfinyl)methyl)phenyl-2-bromo-2-methylpropanoate.

In embodiments, the poly(*p*-arylene vinylene) monomer may be a 1-(chloromethyl)-5-alkoxy-2-alk'oxy-4-((octylsulfinyl)methyl)benzene.

A hydrophobic or hydrophilic 1-(chloromethyl)-5-alkoxy-2-alk'oxy-4-((octylsulfinyl)methyl)benzene is obtained through the choice of one or more hydrophobic or hydrophilic alkoxy substituents, wherein the 5-alkoxy and 2-alk'oxy may be independently chosen.

In embodiments, the vinyl monomer may be a hydrophilic 2-hydroxyethyl acrylate, ethylene glycol monomethyl ether methacrylate, or n-(2-hydroxypropyl) methacrylamide, or a hydrophobic polystyrene.

The invention will now be described by a detailed description of several embodiments of the invention. It is clear that other embodiments of the invention can be configured according to the knowledge of the person skilled in the art without departing from the true technical teaching of the invention, the invention being limited only by the terms of the appended claims.

<u>Example 1a: Synthesis of amphiphilic block copolymers, wherein a hydrophobic block comprises the conjugated polymer</u>

PPV-*b*-PEGMA, PPV-*b*-PHEA and PPV-*b*-PHPMA amphiphilic block copolymers in accordance with embodiments of the present invention were synthesized. In these polymers, the PPV conjugated polymer formed a hydrophobic block due to the selected side chains and the PEGMA, PHEA or PHPMA formed a hydrophilic block.

We now refer to Fig. 2. In a first part, a PPV precursor monomer, 1-(chloromethyl)-5-((3,7-dimethyloctyl)oxy)-2-methoxy-4-((octylsulfinyl)methyl)benzene 7 was synthesized in a number of steps via a sulfinyl precursor route.

3,7-dimethyloctyl-4-methylbenzene sulfonate (2): A mixture of 3,7-dimethyloctanol (1) (115.7 g, 0.732 mol, 1 equiv.) and p-toluenesulfonyl chloride (143 g, 0.732 mol, 1 equiv.) in CH_2Cl_2 (500 mL) was cooled with a mixture of $CHCl_3$ and liquid nitrogen while KOH (166 g, 2.981 mol, 4 equiv.) was added under nitrogen atmosphere. After addition of the base—enabling temperatures below 5°C—the reaction was stirred for 3 h at 0 °C. The reaction was quenched with ice water (500 mL), extracted with CH_2Cl_2 (3 × 250 mL) and the organic layer was dried over anhydrous $MgSO_4$. After filtration, evaporation of the solvent under reduced pressure gave the crude product (2) as a clear oil. No purification was needed (214.80 g, 93.9%). ^{1}H-NMR ($CDCl_3$): δ = 7.74 (m, 2H); 7.30 (m, 2H); 4.01 (m, 2H); 2.39 (s, 3H); 1.59 (m, 1H); 1.42 (m, 3H); 1.06 (m, 6H); 0.79 (m, 9H).

1-((3,7-Dimethyloctyl)oxy)-4-methoxy benzene (3): 4-methoxyphenol (75.45 g, 0.608 mol, 1 equiv.) and NaO*t*Bu (70.49 g, 0.735 mol, 1.21 equiv.) in EtOH (600 mL) was stirred for 1 h at room temperature under nitrogen atmosphere. 3,7-dimethyloctyl-4-methylbenzene sulfonate (2) (208.9 g, 0.67 mol, 1.1 equiv.) was added and the complete mixture was stirred overnight at reflux temperature (80 °C). The reaction was quenched with H_2O (600 mL), extracted with CH_2Cl_2 (3 × 250 mL) and the organic layer was dried over anhydrous $MgSO_4$. After filtration, evaporation of the solvent under reduced pressure gave the crude product (3) as a brown oil. The pure product (3) was obtained by vacuum distillation at 95 °C (120.36 g, 75.1%). ^{1}H-NMR ($CDCl_3$): δ = 6.83 (s, 4H); 3.92 (s, 2H); 3.76 (s, 3H); 1.78 (s, 1H); 1.56 (m, 3H); 1.31 (s, 4H); 1.17 (s, 2H); 0.89 (s, 9H). ^{13}C-NMR ($CDCl_3$): δ = 154.1 (C4); 152.6 (C4); 115.8 (CH); 115.6 (CH); 67.2 (CH_2); 55.8 (CH_3); 39.2 (CH_2); 37,6 (CH_2); 37,1 (CH_2); 29.2 (CH_2); 28.7 (CH_2); 23.2 (CH_3); 15.24 (CH_3). DIP MS (CI, *m/z*): 265 (M+), 220/221 (M^+–OEt), 142/143 (M^+–OPhOMe). FT–IR (ATR): ν = 2995, 2482, 1516, 825, 749 cm^{-1}.

2,5-Bis(chloro-methyl)-1-(3,7-dimethyloctyloxy)-4-methoxybenzene (4): To a stirred mixture of 1-((3,7-Dimethyloctyl)oxy)-4-methoxy benzene (3) (120.63 g, 0.46 mol, 1 equiv.) and *p*-formaldehyde (37.76 g, 1.25 mol, 2.75 equiv.), HCl (37%, 296.60 g, 3.01 mol, 6.6 equiv.) was added drop wise at room temperature under nitrogen atmosphere. Subsequently acetic anhydride (466 g, 4.56 mol, 10 equiv.) was added drop wise, without exceeding a temperature of 70 °C. The solution was stirred at 70 °C for 4 h. After cooling down to room temperature, H_2O (600 mL) was added to the solution. The resulting precipitate was filtered off and redissolved in CH_2Cl_2 (400 mL). Next, the organic solution was dried over anhydrous $MgSO_4$ and filtered. Evaporation of the solvent under reduced pressure gave the crude product as a yellow oil. The pure product (4) was obtained by recrystallization in hexane as white crystals (102.3 g, 61.43%). M_p: 65 °C. ^{1}H-NMR ($CDCl_3$): δ = 6.90 (s, 4H); 4.61 (s, 4H); 4.00 (s, 2H); 3.84 (s, 3H); 1.80 (s, 2H); 1.61 (m, 5H); 1.29 (m, 2H); 1.12 (m, 2H); 0.90 (m 9H); ^{13}C-NMR ($CDCl_3$): δ = 19.8 (CH_3); 22.6 (CH_3); 24.6 (CH_2); 27.9 (CH), 30.2 (CH_2), 36.6 (CH_2), 37.4 (CH_2), 39.2 (CH_2), 41.2 (CH_2), 56.4 (CH_3), 67.9 (CH_2), 113.2 (C4), 114.3 (C4), 127.0 (C4). DIP MS (CI, *m/z*): 361 (M+), 326 (M^+–Cl), 290 (M^+–2Cl); FT–IR (ATR): ν = 2948, 2878, 1977, 1508, 1459, 1401, 885, 881, 735, 690 cm^{-1}.

1,4 –Bis(tetrahydrothiopheniomethyl)xylene dichloride (5): To a stirred mixture of 2,5-Bis(chloro-methyl)-1-(3,7-dimethyloctyloxy)-4-methoxybenzene (4) (65.94 g, 0.18 mol, 1 equiv.) in MeOH (600 mL), tetrahydrothiophene (THT) (80.45 g, 0.91 mol, 5 equiv.) was added. The reaction was allowed to react at room temperature for 3 days. The solution was precipitated in cold acetone (2 L) under heavy stirring and the resulting precipitate was filtered off and washed with cold acetone. After drying under vacuum, the pure product 5 was obtained as a white solid (60.78 g, 60.04%). M_p: 81–83 °C. ^{1}H-NMR (D_2O): δ = 7.10 (s, 2H); 4.41 (s, 4H); 4.08 (m, 4H); 3.80 (s 2H); 3.40 (d, *J* = 5.5 Hz, 8H); 2.22 (m, 8H); 1.74 (m, 2H); 1.55–1.37 (m, 3H); 1.18 (m, 6H); 0.84 (d, *J* = 6.3 Hz, 3H); 0.71 (dd, *J* = 6.6 and 2.9, 6H). ^{13}C-NMR (D_2O): δ = 19.10, (CH_3); 22.07 (CH_3); 24.13 (CH_2); 26.4 (CH); 29.6 (CH); 30.7 (CH_2); 31.3 (CH_2); 37.6 (CH_2); 38.7 (CH_2); 40.9 (CH_2); 43.9 (CH_2); 45.5 (CH_2); 58.6 (CH_3); 70.0 (CH_2); 117.8/118.6 (CH); 121.9/122.4 (C4); 153.7/154.3 (C4). DIP MS (CI, *m/z*): 556 (MH^+), 524 (M^+–Cl), 432 (M^+–THT–Cl), 296 (M^+-2 THT–Cl–CH_2). FT–IR (ATR): ν = 3016, 2926, 2462, 1633, 1514, 1463, 1417, 1317, 1226, 920, 786, 699 cm^{-1}.

(4-(chloromethyl)-2-((3,7-dimethyloctyl)oxy)-5-methoxybenzyl)(octyl)sulfane (6): A mixture of *n*-octanethiol (15.82 g, 0.108 mol, 1 equiv.) and NaO*t*Bu (10.04 g, 0.108 mol, 1 equiv.) in MeOH (240 mL) was stirred at room temperature for 30 min. This mixture was added drop wise to a stirred mixture of 1,4 –Bis(tetrahydrothiopheniomethyl)xylene dichloride (5) (60.0 g, 0.108 mol, 1 equiv.) in MeOH (360 mL) after which it was allowed to react for 3 h at room temperature under nitrogen atmosphere. After evaporation of the solvent under reduced pressure, n-octane (100 mL) was added and evaporated again to remove the THT by azeotropic distillation. This procedure was repeated 5 times. The residue was redissolved in CH_2Cl_2 (350 mL), extracted with a saturated NaCl:H_2O solution (1:10) (3 × 250 mL) and the organic layer was dried over anhydrous $MgSO_4$. After filtration, evaporation of the solvent under reduced pressure gave the crude product 6 as a white solid (49.03 g, 96.9%), which was used without further purification. ^{1}H-NMR ($CDCl_3$): δ = 6.87 (m, 2H); 4.62 (d, *J* = 2.4 Hz, 2H); 3.99 (m, 2H); 3.84 (m, 3H); 3.70 (d, *J* = 3.4 Hz, 2H); 2.45 (t, *J* = 6.8 Hz, 2H); 1.87 (m, 1H); 1.81 (m, 1H); 1.52 (m, 2H); 1.25 (m, 18H); 0.88 (m, 9H).

1-(chloromethyl)-5-((3,7-dimethyloctyl)oxy)-2-methoxy-4-((octylsulfinyl)methyl)benzene (7): To a stirred mixture of (4-(chloromethyl)-2-((3,7-dimethyloctyl)oxy)-5-methoxybenzyl)(octyl)sulfane (6) (49.0 g, 0.104 mol, 1 equiv.) in 1,4-dioxane (800 mL), TeO_2 (2.075 g, 0.013 mol, 0.125 equiv.) and HCl (2 M, 4.9 mL, 0.122 mol, 1.2 equiv.) were added. To start the reaction H_2O_2 (35%, 20.20 g, 0.208 mol, 2 equiv.) was added and the reaction was followed on TLC (hexane/ EtOAc; 6/4). As soon as all (4-(chloromethyl)-2-((3,7-dimethyloctyl)oxy)-5-methoxybenzyl)(octyl)sulfane (6) was consumed, the reaction was quenched with a saturated NaCl-solution:H_2O (1:1; 400 mL). The solution was extracted with CH_2Cl_2 (3 × 300 mL), dried over anhydrous $MgSO_4$ and filtered. Evaporation of the solvent under reduced pressure gave the crude product as a yellow oil. The pure product (7) was obtained by column chromatography (SiO_2, hexane/EtOAc 6/4) (25.48 g, 50.36%). M_p: 109.5–110.5 °C. ^{1}H-NMR ($CDCl_3$): δ = 7.37 (d, *J* = 8.0 Hz, 2H); 7.26 (d, *J* = 8.0 Hz, 2H); 4.55 (s, 2H); 3.91 + 3.93 (dd, J_{AB} = 13.0 Hz, 2H); 2.53 (t, *J* = 8.0 Hz, 2H); 1.70 (m, 2H); 1.36 (m, 2H); 1.23 (m, 8H); 0.84 (t, *J* = 6.8 Hz, 3H); ^{13}C NMR ($CDCl_3$,100 MHz): δ = 14.0 (CH_2); 22.4 (CH_3); 22.5 (CH_3); 28.7 (CH); 28.9 (CH_2); 29.1 (CH); 31.6 (CH_2);

45.6 (CH_2), 51.0 (CH_2); 57.6 (CH_3); 129.1 (C4); 130.2 (C4); 130.3 (C4); 137.5 (C4). DIP MS (CI, *m/z*): 488 (MH^+), 451/453 (M^+–Cl), 325/327 (M^+–S(O)C_8H_{17}), 291 (M^+–Cl–S(O)C_8H_{17}), 174 (S(O)C_8H_{17}); FT–IR (ATR): ν = 2959, 2918, 2847, 1520, 1457, 1405,919, 754, 689 cm^{-1}.

We now refer to Fig. 3. In a second part, the (4-[(methylsulfinyl)methyl]phenyl-2-bromo-2-methylpropanoate) initiator (11) was synthesized in a number of steps.

4-((methylthio)methyl)phenol (9): To a mixture of NaSMe (21% in H_2O, 4.33 g, 12.97 mmol, 1.1 equiv.) in EtOH (8 mL), 4-(chloromethyl)phenyl acetate (8) (2.28 g, 12.36 mmol, 1 equiv.) was added while stirring and further stirred for 1 h at reflux temperature (80°C). A solution of KOH (1.38 g, 24.72 mmol, 2 equiv.) in H_2O (50 mL) was added and again stirred for 1 h at room temperature. The solution was acidified with HCl (2M) to a pH~2. The solution was extracted with diethyl ether (3 x 100 mL) and the organic layer was dried over anhydrous $MgSO_4$. After filtration, evaporation of the solvent under reduced pressure gave the crude product (9) as an orange oil. The pure product (9) was obtained by column chromatography (SiO_2, eluent CH_2Cl_2) as a colorless oil (1.04 g, 54.5 %). ^{1}H-NMR ($CDCl_3$): δ = 7.15 (d, *J* = 8.2 Hz, 2H); 6.76 (d, *J* = 8.5 Hz, 2H); 5.28 (s, 1H); 3.60 (s, 2H); 1.97 (s, 3H). ^{13}C-NMR ($CDCl_3$): δ = 150.3 (C4); 136.9 (C4) ; 130.7 (CH); 115.9 (C4); 38.3 (CH_2); 15.5 (CH_3).

4-((methylthio)methyl)phenyl-2-bromo-2-methylpropanoate (10): 2-bromo-2-methylpropanoylbromide (1 mL, 8.77 mmol, 1.3 equiv) was added drop wise to a stirred mixture of 4-((methylthio)methyl)phenol (9) (1.04 g, 6.74 mmol, 1 equiv.), CH_2Cl_2 (50 mL) and pyridine (1 mL, 13.49 mmol, 2 equiv.) at 0°C. The solution was stirred overnight at room temperature. The reaction was quenched with water (50 mL), extracted with CH_2Cl_2 (3 x 100 mL), dried over anhydrous $MgSO_4$ and filtered. Evaporation of the solvent under reduced pressure gave the crude product (10) as a yellow oil (2.51 g) and the product was used without further purification. ^{1}H-NMR ($CDCl_3$): δ = 7.32 (d, *J* = 8.6 Hz, 2H); 7.06 (d, *J* = 8.6 Hz, 2H); 2.05 (s, 6H); 1.98 (s, 5H). ^{13}C-NMR ($CDCl_3$): δ = 170.2 (C4); 149.6 (C4); 136.2 (C4); 129.9 (CH); 121.0 (CH); 55.2 (C4); 37.5 (CH2); 30.6 (CH3); 14.9 (CH3).

4-((methylsulfinyl)methyl)phenyl-2-bromo-2-methylpropanoate (11): To a stirred mixture of 4-((methylthio)methyl)phenyl-2-bromo-2-methylpropanoate (10) (2.51g, 8.31 mmol, 1 equiv.) in 1,4-dioxane (50 mL), TeO_2 (0.26 g, 1.66 mmol, 1/5 equiv.) and HCl (2M, 0.40 mL) were added. To start the reaction H_2O_2 (35%; 1.42 mL, 16.62 mmol, 2 equiv.) was added. The reaction was followed on TLC ($CHCl_3$/petroleum ether; 6/4). As soon as all 4-((methylthio)methyl)phenyl-2-bromo-2-methylpropanoate (10) was consumed, the reaction was quenched with a saturated NaCl-solution: H_2O (1:1; 50 mL). The solution was extracted with CH_2Cl_2 (3 x 100 mL), dried over anhydrous $MgSO_4$ and filtered. Evaporation of the solvent under reduced pressure gave the crude product (11) as a yellow oil. The pure product (11) was obtained by crystallization in hexane/EtOAc (3/1; 3 mL/1 g product) as white crystals. (1.23 g, 31.23 %). Mp: 79°C. ^{1}H-NMR ($CDCl_3$): δ = 7.33 (d, *J* = 8.2 Hz, 2H); 7.16 (d, *J* = 8.6 Hz, 2H); 2.46 (s, 2H); 2.05 (s, 6H). ^{13}C-NMR ($CDCl_3$): δ = 162.7 (C4); 143.5 (C4); 123.8 (CH); 120.2 (C4); 114.3 (CH); 51.9 (CH2); 47.8 (C4); 29.8 (CH3); 23.2 (CH3). DIP MS (CI, *m/z*): 319/321 (MH^+), 255/257 (M^+-S(O)Me). FT-IR (NaCl): ν = 3005, 2977, 2925, 1751, 1504, 1464, 1267, 1210, 1035, 1017 cm^{-1}.

We now refer to Fig. 4. In a third part, the different blocks of the amphiphilic block copolymers were synthesized to the initiator (11).

First, the PPV precursor monomer (7) was polymerized with the first initiating moiety of the initiator (11) to obtain a precursor of the first block. All glassware was dried overnight in a drying oven at 110°C and flame-dried under vacuum. PPV precursor monomer (7) (0.119 g, 0.25 mmol, 1 equiv.) and initiator (11) (0.015 g, 0.05 mmol, 0.2 equiv.) (flushed 3 times with N_2 and vacuum) were dissolved in dry THF (4.67 mL) under N_2 atmosphere at 0°C. While stirring under N_2 atmosphere, lithium bis(trimethylsilyl) amide (LHMDS; 97%, Sigma Aldrich) (1M in THF; 0.33 mL, 0.33 mmol, 1.3 equiv.) was added to start the reaction. After 15 minutes the reaction was quenched with HCl (37%; 0.2 mL) and the solution was poured into H_2O (10 mL). After extraction with CH_2Cl_2 (3 x 50 mL) and evaporation, the product (12) was isolated as a bright yellow sticky oil.

Subsequently, a (meth)acrylate ester precursor or a (meth)acrylamide precursor was polymerized to the second initiating moiety of the initiator (11) through a single electron transfer living radical polymerization (SET-LRP), to obtain a second block, and a thermal elimination was finally performed to obtain the PPV first block. Following this scheme, 3 amphiphilic block copolymers were synthesized: PPV-*b*-PEGMA, PPV-*b*-PHEA and PPV-*b*-PHPMA.

PPV-b-PEGMA block copolymer (17): For the block copolymer synthesis with ethylene glycol methyl ether methacrylate (EGMA) the non-purified precursor polymer (12) was used. A Schlenk tube was filled with tris[2-(dimethylamino) ethyl]amine (Me_6TREN; 5.3 µL, 19.8 µmol, 1 equiv.), EGMA (0.128 g, 0.89 mmol, 45 equiv.) and precursor polymer (12) (M_n = 5 100 g·mol^{-1}, 0.1 g, 19.8 µmol, 1 equiv.) dissolved in DMF (1 mL). The Schlenk tube was subjected to five freeze pump thaw cycles, after which it was transferred into the glove box. Cu(0) (1.26 mg, 19.8 µmol; 1 equiv.) was added to the Schlenk tube to start the reaction. The mixture was stirred for 4 hrs at room temperature, after which the content was poured into an aluminum tray to evaporate the solvent. The precursor block copolymer (14) was filtered over a column of basic alumina to remove all copper and subsequently all solvent was evaporated. In order to obtain the conjugated block copolymer (17), toluene (10 mL) was added to precursor block copolymer (14) and stirred for 3 hrs under N_2 atmosphere at reflux temperature (110°C). When cooled down to room temperature. PPV-*b*-P(EGMA) block copolymer (17) was isolated as a red / orange viscous solution after precipitation in MeOH / H_2O (4/1) mixture and filtration. SEC (DMAc): M_n^{app} = 9 500 g·mol^{-1}, M_w^{app} = 12 400, $Đ$ = 1.3. ATR FT-IR: 2955, 2927, 2365, 1677, 1559, 1479, 1301, 1215, 1097, 1042, 747, 672, 629, 544 cm^{-1}. UV-Vis ($CHCl_3$): $\lambda_{max,ex}$ = 418 nm. Fluorescence ($CHCl_3$): $\lambda_{max,em}$ = 506 nm.

PPV-b-PHEA block copolymer (18): The synthesis was similar to synthesizing PPV-*b*-PEGMA (17), however, HEA and a reaction time of 2 hrs were used. The conjugated block copolymer (18) was again obtained after thermal elimination of precursor block copolymer (15). SEC (DMAc): M_n^{app} = 16 100 g·mol^{-1}, M_w^{app} = 22 900, $Đ$ = 1.4. ATR FT-IR: 3397, 2945, 2476, 2349, 1717, 1441, 1392, 1253, 1162, 1064, 1021, 893, 838, 774, 624, 516 cm^{-1}. UV-Vis (DMSO): $\lambda_{max,ex}$ = 438 nm. Fluorescence (DMSO): $\lambda_{max,em}$ = 512 nm.

PPV-b-PHPMA block copolymer (19): The synthesis was similar to PPV-*b*-PEGMA (17), however, HPMA and a reaction time of 3 days were used in combination with $CuCl_2$ (11.6 mg, 87 µmol,

1 equiv.). The conjugated block copolymer (19) was again obtained after thermal elimination of precursor block copolymer (16). SEC (DMAc): M_n^{app} = 5 100 g·mol^{-1}, M_w^{app} = 6 100, $Đ$ = 1.2. ATR FT-IR: 3305, 2983, 2854, 2355, 1726, 1656, 1602, 1441, 1409, 1376, 1333, 1210, 1124, 1032, 903, 823, 646, 559 cm^{-1}. UV-Vis (DMSO): $\lambda_{max,ex}$ = 466nm. Fluorescence (DMSO): $\lambda_{max,em}$ = 546 nm.

Example 1b: Synthesis of amphiphilic block copolymers, wherein a hydrophilic block comprises the conjugated polymer

Alternative to example 1a, the conjugated polymer may also be part of a hydrophilic block. As an example to this effect, PPV-*b*-PS was synthesized. Herein, the PPV conjugated polymer formed a hydrophilic block due to the selected side chains whereas the PS formed a hydrophobic block.

We now refer to Fig. 5. In a first part, a PPV precursor monomer, 6-(5-chloromethyl-4-methoxy-2-octylsulfinylmethylphenoxy)-hexanoic acid methyl ester (CPM; 25) was synthesized in a number of steps via a sulfinyl precursor route.

6-(4-methoxy-phenoxy)-hexanoic acid ethyl ester (21): A mixture of 4-methoxy-phenol (20) (93.5 g, 0.75 mol, 1 equiv.) and NaO*t*Bu (86.6 g, 0.90 mol, 1.2 equiv.) in EtOH (500 mL) was stirred for 1 h at room temperature under nitrogen atmosphere. A solution of ethyl-6-bromohexanoate (200 g, 0.90 mol, 1.2 equiv.) and NaI (3.4 g, 0.02 mol, 1.2 equiv.) in EtOH (200 mL) was added and the complete mixture was stirred for 4 h at reflux temperature (80 °C). The reaction was quenched with H_2O (400 mL), extracted with CH_2Cl_2 (3 × 100 mL) and the organic layer was dried over anhydrous $MgSO_4$. After filtration, evaporation of the solvent under reduced pressure gave the crude product as an orange oil. The pure product **9** was obtained by recrystallization in MeOH as white crystals (131.61 g, 65.70%). M_p: 30 °C. ^{1}H-NMR ($CDCl_3$): δ = 6.80 (s, 4H); 4.10 (q, *J* = 7.3 Hz, 2H); 3.88 (t, *J* = 6.4 Hz, 2H); 3.74 (s, 3H); 2.30 (t, *J* = 7.6 Hz, 2H) ; 1.74 (m, 2H); 1.68 (m, 2H); 1.47 (m, 2H); 1.23 (t, *J* = 7.2 Hz, 3H). ^{13}C-NMR ($CDCl_3$): δ = 173.74 (C4); 153.65 (C4); 153.13 (C4); 115.35 (CH); 114.56 (CH); 68.24 (CH_2); 60.24 (CH_2); 55.70 (CH_3); 34.25 (CH_2); 29.04 (CH_2); 25.65 (CH_2); 24.72 (CH_2); 14.24(CH_3). DIP MS (CI, *m/z*): 266 (M+), 221/222 (M^+–OEt), 143/144 (M^+–OPhOMe), 124/125 (M^+–C_5H_{10}COOEt). FT–IR (ATR): ν = 2938, 2479, 2000, 1866, 1732, 1506, 1476, 1292, 1233, 1160, 1112, 1033, 825, 742 cm^{-1}.

6-(2,5-bis-chloromethyl-4-methoxy-phenoxy)hexanoic acid (22): To a stirred mixture of 21 (131.6 g, 0.49 mol, 1 equiv.) and *p*-formaldehyde (40.9 g, 1.35 mol, 2.75 equiv.), HCl (37%, 110.1 g, 3.26 mol, 6.6 equiv.) was added drop wise at 0 °C under nitrogen atmosphere. Subsequently acetic anhydride (503 g, 4.94 mol, 10 equiv.) was added drop wise, without exceeding a temperature of 70 °C. The solution was stirred at 60 °C for 3 h. After cooling down to room temperature, H_2O (400 mL) was added to the solution. The resulting precipitate was filtered off and redissolved in CH_2Cl_2 (200 mL). Next, the organic solution was dried over anhydrous $MgSO_4$ and filtered. Evaporation of the solvent under reduced pressure gave the crude product as a yellow oil. The pure product 22 was obtained by recrystallization in EtOAc as white crystals (71.7 g, 41.74%). M_p: 99 °C. ^{1}H-NMR ($CDCl_3$): δ = 6.90 (d, *J* = 3.9 Hz, 2H); 4.60 (d, *J* = 3.5 Hz, 4H); 3.98 (t, *J* = 6.2 Hz, 2H); 3.83 (s, 3H); 2.39 (t, *J* = 7.4 Hz, 2H); 1.82 (m, 2H); 1.72 (m, 2H); 1.56 (m, 2H). ^{13}C-NMR ($CDCl_3$): δ =171.95 (C4); 151.65 (C4); 151.13 (C4); 127.67 (C4); 127.41 (C4); 114.98 (CH); 113.90 (CH); 69.24 (CH_2); 61.06 (CH_2); 58.80 (CH_3); 41.93 (CH_2); 41.85 (CH_2); 34.53

(CH_2); 29.54 (CH_2); 26.14 (CH_2); 24.93 (CH_2); 21.62 (CH_2); 14.75 (CH_3). DIP MS (CI, *m/z*): 334 (M+), 299 (M^+–Cl), 263 (M^+–2Cl), 220/222 (M^+–$C_5H_{10}COOH$). FT–IR (ATR): ν = 2938, 2875, 1971, 1724, 1687, 1512, 1463, 1410, 1312, 1220, 1141, 1033, 875, 871, 732, 686 cm^{-1}.

6-(2,5-bis-chloroemethyl-4-methoxy-phenoxy) hexanoic acid methyl ester (23): To a stirred mixture of 22 (71.7 g, 0.22mol, 1 equiv.) in MeOH (600 mL), tetrahydrothiophene (THT) (99.8 g, 1.1 mol, 5 equiv.) was added. The reaction was allowed to react at 50 °C for 4 days. The solution was precipitated in cold diethyl ether (1 L) under heavy stirring and the resulting precipitate was filtered off and washed with cold diethyl ether. After drying under vacuum, the pure product 23 was obtained as a white solid (40.0 g; 33.85%). M_p: 119 °C. ^{1}H-NMR (D_2O): δ = 7.08 (s, 1H); 7.06 (s, 1H); 4.41 (s, 2H); 4.40 (s, 2H); 4.01 (t, *J* = 6.3 Hz, 2H); 3.79 (s, 3H); 3.56 (s, 3H); 3.40 (m, 8H); 2.32 (t, *J* = 6.3 Hz, 2H); 2.24 (m, 8H); 1.75 (m, 2H); 1.59 (m, 4H); 1.42 (m, 2H). ^{13}C-NMR ($CDCl_3$): δ = 179.43 (C4); 178.03 (C4); 152.56 (C4); 152.54 (C4); 151.89 (C4); 151.86 (C4); 120.40 (CH); 120.33 (CH); 116.78 (CH); 116.03 (CH); 69.78 (CH_2); 56.94 (CH_3); 52.75 (CH_3); 49.53 (CH_2); 45.47 (CH_2); 43.83 (CH_2); 43.77 (CH_2); 42.18 (CH_2); 34.34 (CH_2); 34.20 (CH_2); 29.08 (CH_2); 28.70 (CH_2); 25.64 (CH_2); 24.69 (CH_2). DIP MS (CI, *m/z*): 525 (MH^+), 493 (M^+–Cl), 401 (M^+–THT–Cl), 265 (M^+–2 THT–Cl–CH_2). FT–IR (ATR): ν = 3004, 2942, 2879, 2122, 1732, 1510, 1449, 1399, 1313, 1225, 1033, 909, 773, 702 cm^{-1}.

6-(5-chloromethyl-4-methoxy-2-octylsulfanylmethyl-phenoxy)hexanoic acid methyl ester (24): A mixture of *n*-octanethiol (12.31 g, 83.9 mmol, 1.1 equiv.) and NaO*t*Bu (8.1 g, 76.3 mmol, 1 equiv.) in MeOH (300 mL) was stirred at room temperature for 30 min. This mixture was added drop wise to a stirred mixture of 23 (40.0 g, 76.3 mmol, 1 equiv.) in MeOH (700 mL) after which it was allowed to react for 2 h at room temperature under N_2 atmosphere. After evaporation of the solvent under reduced pressure, *n*-octane (100 mL) was added and evaporated again to remove the THT. This procedure was repeated 5 times. The residue was redissolved in CH_2Cl_2 (350 mL), extracted with a saturated NaCl-solution:H_2O (1:10) (3 × 250 mL) and the organic layer was dried over anhydrous $MgSO_4$. After filtration, evaporation of the solvent under reduced pressure gave the crude product 24 as a white solid (26.0 g, 67.5%) and used without further purification. ^{1}H-NMR ($CDCl_3$): δ = 6.85 (m, 2H); 4.61 (s, 1H); 4.60 (s, 1H); 3.94 (m, 2H); 3.80 (m, 3H); 3.69 (d, *J* = 3.5 Hz, 2H); 3.65 (s, 3H); 2.45 (t, *J* = 6.8 Hz, 2H); 2.33 (t, *J* = 7.4 Hz, 2H); 1.79 (m, 2H); 1.65 (m, 2H); 1.52 (m, 2H); 1.24 (m, 12 H); 0.85 (m, 3H).

6-(5-chloromethyl-4-methoxy-2-octylsulfinylmethyl-phenoxy)-hexanoic acid methyl ester (25): To a stirred mixture of 24 (26.0 g, 56.6 mmol, 1 equiv.) in 1,4-dioxane (350 mL), TeO_2 (1.1 g, 7.1 mmol, 1/8 equiv.) and HCl (1 M, 10 mL, 1.2 equiv.) were added. To start the reaction H_2O_2 (35%; 11.01 g 113.3 mmol, 2 equiv.) was added and the reaction was followed on TLC (hexane/ EtOAc; 5/5). As soon as all 24 was consumed, the reaction was quenched with a saturated Na_2SO_3-solution:H_2O (1:1; 400 mL). The solution was extracted with CH_2Cl_2 (3 × 300 mL), dried over anhydrous $MgSO_4$ and filtered. Evaporation of the solvent under reduced pressure gave the crude product as a yellow oil. The pure product 25 was obtained by column chromatography (SiO_2, hexane/EtOAc 5/5)(45.97 mmol, 21.80 g, 81.22%). M_p: 35 °C. ^{1}H-NMR ($CDCl_3$): δ = 6.76 (s, 1H); 6.75 (s, 1H); 4.44 (s, 2H); 3.63 (d, *J* = 7 Hz, 4H); 3.48 (s, 3H); 3.47 (s, 3H); 2.45 (t, *J* = 7.4 Hz, 2H); 2.15 (t, *J* = 7.4 Hz, 2H); 1.65–1.45 (m, 6H); 1.32–1.05 (m, 12H); 0.69 (t, *J* = 6.6 Hz, 3H). ^{13}C-NMR ($CDCl_3$): δ = 174.55 (C4); 174.42 (C4); 151.79 (C4); 151.54 (C4);

127.18 (C4); 120.43 (C4); 115.23 (CH); 113.44 (CH); 69.17 (CH_2); 56.74 (CH_3); 53.48 (CH_2); 52.03 (CH_2); 51.90 (CH_3); 41.93 (CH_2); 41.85 (CH_2); 34.34 (CH_2); 32.25 (CH_2); 29.54 (CH_2); 29.39 (CH_2); 26.16 (CH_2); 23.13 (CH_2); 14.61 (CH_3). DIP MS (CI, *m/z*): 475/477 (MH^+), 439/441 (M^+–Cl), 313/315 (M^+–S(O)C_8H_{17}), 279 (M^+–Cl–S(O)C_8H_{17}), 162/163 (S(O)C_8H_{17}). FT–IR (ATR): ν = 3003, 2942, 2881, 2409, 1732, 1545, 1510, 1447, 1400, 1313, 1220, 1165, 1104, 1034, 909, 774, 686 cm^{-1}.

We now refer to Fig. 3. In a second part, the (4-[(methylsulfinyl)methyl] phenyl-2-bromo-2-methylpropanoate) initiator (11) was synthesized in the same way as described in example 1.a.

We now refer to Fig. 6. In a third part, the PPV block of the amphiphilic block copolymers was synthesized to the initiator (11). First, the PPV precursor monomer (25) was polymerized with the first initiating moiety of the initiator (11) to obtain a precursor of the first block, a thermal elimination was then performed and a finally a hydrolysis of the PPV side chains to obtain the final PPV first block.

All glassware was dried overnight in a drying oven at 110 °C and flamed under vacuum prior to use. The precursor monomer (25) (0.2 g, 0.7 mmol) and the given amount of initiator (11) were dissolved in dry THF providing a precursor monomer concentration of 0.05 M and brought at 0 °C under nitrogen atmosphere. The polymerization was started by adding 1.2 equivalents of LHMDS (1M in THF) by syringe. After 1 h, the reaction mixture was precipitated in water, neutralized with 1.0 M HCl, extracted with CH_2Cl_2 and analyzed without further purification.

The precursor polymer (26) was then dissolved in toluene (10 mL) and heated at 110 °C for 3 h. After cooling down, the polymer (27) was precipitated in cold methanol (100 mL) and filtered on a Teflon® filter. The polymer (27) was obtained as a red powder.

A solution of eliminated polymer (27) (0.06 g, 0.19 mmol ester functionalities) and dioxane (20 mL) was heated to reflux temperature after which a solution of KO*t*Bu (0.22 g, 1.94 mmol) in water (1 mL) was added at reflux temperature. After stirring at reflux temperature for 4 hrs, CH_2Cl_2 (150 mL) was added to the reaction mixture and extracted with 1M HCl-solution (100 mL). The organic layer was evaporated under reduced pressure giving the hydrophilic polymer block (28). The resulting red polymer (28) was dried at room temperature under reduced pressure. FT-IR (ATR): ν = 3502, 2960, 2930, 2857, 1733, 1500, 1460, 1413, 1383, 1356, 1258, 1205, 1094, 1030, 968, 860, 800 cm^{-1}.

We now refer to Fig. 7. In a fourth part, a polystyrene (PS) precursor was polymerized to the second initiating moiety of the initiator (11) through a single electron transfer living radical polymerization, to obtain a second block. The synthesis was similar to synthesizing PPV-*b*-PEGMA (17), however, styrene and a reaction time of 6 h was used to obtain the PPV-*b*-PS block copolymer (29).

Example 1c: Synthesis of amphiphilic block copolymers, using different conjugated polymers

In order to further illustrate the versatile nature of the block copolymers in accordance with the present invention, examples of conjugated homopolymer synthesis with a bromine-functionality are given. These conjugated homopolymers with bromine functionality may be seen as alternatives to the PPV(-precursor)-initiator intermediates (12, 28) with a similar bromine functionality, described in the examples

1.a en 1.b. These bromine groups can subsequently be reinitiated by using SET-LRP reaction conditions, again similar to described in examples 1.a en 1.b, leading to block copolymer formation of different types of conjugated polymers. Similar reaction conditions will be applied as for PPV-*b*-PEGMA.

We now refer to Fig. 8. In a first illustration, a bromine functionalized polythiophene monomer (32) is synthesized. This monomer (32) may be polymerized according to procedures well known to the person skilled in the art.

3-[2-(pentyloxy)ethyl]thiophene (31): 2-(thiophen-3-yl)ethanol (30) (5.0 g, 39 mmol) was dissolved in DMSO (150 mL), NaH (60% dispersion in mineral oil, 3.12 g, 78 mmol) was added and this suspension was stirred at room temperature for 30 min. 1-Bromopentane (11.8 g, 78 mmol) was added drop wise and the mixture was stirred for 8 h at room temperature. The reaction was quenched by the addition of a saturated NH_4Cl solution (50 mL), water was added and the reaction mixture was extracted several times with diethyl ether. The organic layer was dried with $MgSO_4$, filtered and evaporated under reduced pressure. The residue was purified on a silica column with an eluent gradient from pure *n*-hexane to 75/25 *n*-hexane/DCM. The pure fractions were collected and evaporated, yielding a colorless oil (7.24 g, 94%). ^{1}H NMR (300 MHz, $CDCl_3$): δ = 7.23 (dd, *J* = 5.1 and 3.0 Hz, 1H), 7.02–6.99 (m, 1H), 6.97 (dd, *J* = 4.9 and 1.3 Hz, 1H), 3.62 (t, *J* = 7.2 Hz, 2H), 3.43 (t, *J* = 6.9 Hz, 2H), 2.90 (t, *J* = 7.0 Hz, 2H), 1.58 (q, *J* = 6.9 Hz, 2H), 1.36–1.24 (m, 4H), 0.92–0.86 (m, 3H); ^{13}C NMR (75 MHz, $CDCl_3$): δ = 140.0, 129.2 (CH), 125.8 (CH), 121.7 (CH), 71.8 (CH_2), 71.6 (CH_2), 31.4 (CH_2), 30.1 (CH_2), 29.0 (CH_2), 23.2 (CH_2), 14.7 (CH_3); MS (EI): *m/z* = 198 (M^+).

2,5-dibromo-3-[2-(pentyloxy)ethyl]thiophene (32): 3-(2-(pentyloxy)ethyl)thiophene (31) (7.24 g, 36.5 mmol) was dissolved in THF (150 mL), cooled to 0 °C and protected from light. NBS (14.3 g, 80.3 mmol, 2.3 equiv) was added portion wise and the mixture was stirred further at rt for 8 h in the absence of light. The reaction mixture was quenched by pouring it into an ice-cold solution of 1 M NaOH and the product was extracted with Et_2O. The organic layer was dried with $MgSO_4$, filtered and the solvent was evaporated under reduced pressure. The residue was purified on a silica plug with 50/50 hexanes/CH_2Cl_2 as the eluent. The pure fractions were collected and the solvent was removed under reduced pressure affording a colourless oil (12.0 g, 92%). ^{1}H NMR (300 MHz, $CDCl_3$): δ = 6.84 (s, 1H), 3.52 (t, *J* = 6.9 Hz, 2H) 3.39 (t, *J* = 6.7 Hz, 2H), 2.76 (t, *J* = 6.7 Hz, 2H), 1.54 (q, *J* = 6.9 Hz, 2H), 1.33–1.21 (m, 4H), 0.87 (t, *J* = 6.9 Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$): δ = 140.4, 132.2 (CH), 111.1, 109.6, 71.8 (CH_2), 70.1 (CH_2), 30.8 (CH_2), 30.1 (CH_2), 29.1 (CH_2), 23.3 (CH_2), 14.8 (CH_2); MS (EI): *m/z* = 354/356/358 (M^+).

In a second illustration, a bromine functionalized poly(p-arylene ethynylene) (40) is synthesized.

We now refer to Fig. 9. As a first building block combination, both monomers (37 and 39) needed for the Sonogashira polymerization reaction were synthesized via similar pathways. The first step involves a Williamson ether synthesis in which *p*-hydroquinone (33) was dialkylated with 1,8-dibromooctane or bromooctane, respectively. A large excess of 1,8-dibromooctane was used to prevent two-fold reaction on one alkyl chain and potassium carbonate was found to give better yields than sodium hydroxide or sodium

hydride (in ethanol). Iodination of dialkylated precursors 34 and 38 with iodine and potassium iodate in acidic medium gave products 35 and 39 in acceptable yields (62 and 77%, respectively). The alkyne entities on compound 36 were then introduced via a Sonogashira reaction with trimethylsilylacetylene (TMSA) in the presence of triethylamine. $Pd(PPh_3)Cl_2$ was used as the catalyst and CuI as co-catalyst. From ^1H NMR analysis it was observed that some of the bromine atoms on the alkyl side chains were replaced by iodine during this reaction, leading to an extra triplet at 3.18 ppm. The amount of iodinated product depends on the amount of co-catalyst causing the halogen-halogen exchange. Because iodine is anyway easier substituted later on, this exchange does not pose any problems for the further reaction sequence. The product mixture was used as starting material for the alkyne deprotection reaction. Tetra-*n*-butylammonium fluoride (TBAF) was initially used for this purpose. However, besides the desired reaction, also halogen exchange occurred leading to fluorinated alkyl side chains. Because this side reaction poses reactivity problems for the azidation to follow, deprotection was performed with potassium hydroxide in a methanol/THF mixture at 0 °C, affording 2,5-diethynylbenzene derivative 36 in ~65% yield (Sonogashira + deprotection step, mixture of iodo- and bromo-substituted side chains). The azide groups were finally introduced by reaction of **35** with sodium azide in DMSO, yielding monomer 37 in over 80% yield.

We now refer to Fig. 10. For a second possible building block combination, monomers 40 and 41 could readily be synthesized from products 35 and 39, respectively. The azide functionalities were introduced on precursor 35 in a highly efficient manner (96% yield) using the same reaction conditions as applied for compound 36, and the Sonogashira reaction on 2,5-diiodobenzene derivative 39 was conducted in the same way as mentioned above, in this case with TBAF as deprotection agent, yielding 2,5-diethynylbenzene monomer 41 in 72% yield.

For the pre-polymerization functionalization route, monomers 40 and 41, with the bromine groups on 2,5-diiodobenzene monomer 40, were copolymerized via Sonogashira polymerization with $Pd(PPh_3)_2Cl_2$ as a catalyst and CuI as co-catalyst. The reaction was performed in a toluene/diisopropylamine (DIPA) mixture at 70 °C. The same reaction was also done starting from monomers 37 and 39, in this case with the bromine functions on the 2,5-diethynylbenzene building block. None of the polymers were purified by soxhlet extraction because elevated temperatures can cause cross-linking via thermally activated Huisgen cycloaddition. The polymers were precipitated in methanol and subsequently in acetone to remove catalyst residues and low molar mass fractions. The polymerization of monomers 35 and 37 yielded bromine-functionalized copolymer with an M_n of 12.5 kg/mol and a polydispersity of 2.1, whereas combination of 34 and 39 yielded copolymer with an M_n of 19.3 kg/mol and a polydispersity of 2.1

Example 2: Self-assembly and characteristics of amphiphilic block copolymer micelles

In order to obtain the micelles, 2 mg of the block copolymer (17, 18 or 19) was dissolved in 0.4 mL of DMF. The solution was placed on a stir plate with a high stirring rate at room temperature. Deionized water (3.6 mL) was added dropwise to the solution with a flow rate of 0.2 $mL \cdot h^{-1}$, leading to a

total concentration of 0.5 mg·mL^{-1}. Afterwards, in order to remove the DMF, the solution was placed in a dialysis membrane with pore size M_w < 3 500 g·mol^{-1} and dialyzed against deionized water for 48 hrs.

Different characteristics of the obtained micelles are displayed in the table below: intensity (I_{mean}), volume (V_{mean}) and number (N_{mean}) average sizes and the dispersity (Đ) as obtained through dynamic light scattering (DLS), the average size (D_{50}) as obtained through transmission electron microscopy (TEM), the maximum wavelength (λ_{max}) of the absorption (Abs) and emission (Em) spectrum and the zeta potential as obtained through DLS. For comparison, the absorption and emission maxima of the free polymers in solution ($CHCl_3$ for PPV-b-PEGMA; DMSO for PPV-b-PHEA and PPV-b-PHPMA) are repeated as well.

Polymer content	**Addition flow rate**	**DLS nm**				**TEM nm**	**λ_{max} nm**		**Zeta potential**
mg·ml^{-1}	**ml·h^{-1}**	***I*$_{mean}$**	***V*$_{mean}$**	***N*$_{mean}$**	***Đ***	***D*$_{50}$**	***Abs***	***Em***	**mV**
PPV-*b*-PEGMA									
-	-	-	-	-	-	-	418	506	-
0.5	0.2	119.3	109.2	94.5	0.197	22 ± 2	361	464	-28.9
0.5	0.1	127.8	108.6	82.6	0.096	11 ± 3	361	470	-33
PPV-*b*-PHEA									
-	-	-	-	-	-	-	438	512	-
0.5	0.2	183.5	182.5	156.7	0.337	30 ± 15	389	491	-35.7
PPV-*b*-PHPMA									
-	-	-	-	-	-	-	466	512	-
0.5	0.2	346.4	341.7	158.3	0.363	50 ± 5	421	495	-38.7

The obtained micelles were shown to be stable for a period of at least 1.5 years.

We now refer to Fig. 12. The cytotoxicity of the amphiphilic block copolymer micelles inside living cells was investigated using the protocol described hereafter. No cytotoxicity was apparent for PPV-*b*-PEGMA (a) and PPV-*b*-PHEA (b), while a cytotoxicity corresponding to a half maximal inhibitory concentration of 0,599 µM was observed for PPV-*b*-PHPMA (c). However, since HPMA is not toxic, it is believed that this observed cytotoxicity may not be due to the PPV-*b*-PHPMA itself, but rather due to a remnant of copper which was used during its synthesis.

For the living cells, human pancreatic carcinoma AsPC-1 cells were cultured in T25 cell culture flask with 5 % CO_2 at 37 °C. The culture medium was composed of RPMI1640 medium (Thermo Fisher Scientific, Australia) supplemented with 10% fetal bovine serum (Bovogen Biologicals, Australia), 100 U/mL penicillin (Sigma-aldrich, Australia), 100 µg/mL streptomycin (Sigma-aldrich, Australia) and 1× GlutaMAX™ (Gibco, Thermo Fisher Scientific, Australia). After the cells reached confluence, the cells were washed with phosphate buffered saline (PBS) and detached by trypsin/EDTA treatment (Sigma-

aldrich, Australia). The cells were collected, centrifuged and resuspended in the culture medium for the further experiments.

The cytotoxicity of the micelles was subsequently measured using a WST-1 assay (Abam, Australia). AsPC-1 suspension was seeded in 96-well cell culture plates at a density of 4.000 cells per well and cultured with 100 µL cell culture medium at 37 °C for 1 day. The micelles were sterilized by passing through a sterile 0.45 µm membrane and serially diluted with sterile MilliQ water. Then the micelles were added into the plate at 100 µL per well along with 100 µL 2× concentrated cell culture medium. After incubation for 3 days with micelles, 10 µL WST-1 per well was added into the cell culture medium. The plates were then incubated for an additional 2 hours at 37 °C. After incubation, 100 µL of solution was taken out in a new 96-well plate and the absorbance of the samples against the background control on a Benchmark Microplate Reader (Bio-Rad) was obtained at a wavelength of 440 nm with a reference wavelength of 650 nm. Sterile MilliQ water was used instead of micelle solution as a control. All cytotoxicity data are reported as mean ± standard deviation (n = 4).

The cellular uptake of the AsPC-1 cells, incubated for 72 h, of PPV-*b*-PEGMA, PPV-*b*-PHEA and PPV-*b*-PHPMA amphiphilic block copolymer micelles was investigated using confocal fluorescence and differential interference contrast (DIC) microscopy; following the protocol described below. The confocal fluorescence of the amphiphilic block copolymers, the fluorescence of the cell lysosomes, the DIC of the corresponding cell sample and an overlay of all 3 were imaged. As could be observed in all 3 cases, the amphiphilic block copolymers were taken up well within the cells, i.e. they were not exclusively present near the edges, and their fluorescence generally corresponded to the location of the lysosomes. Furthermore, as the micelles in their aqueous medium, prior to being contacted to the cell culture, do not display a very high fluorescence, the presence of a strong amphiphilic block copolymer fluorescence inside the cells points towards a spontaneous breaking up of the micelles inside the cell. This is a very interesting property as it means that any loaded compound inside the micelles will be readily released once inside the cell.

In order to measure this cellular uptake, AsPC-1 cells were seeded in 35 mm Fluoro-dishes (0.5 × 105 cells per dish) and incubated for 3 days at 37 °C and 5 % CO_2. The micelles were sterilised by passing through a sterile 0.45 µm membrane and loaded to the cells at a concentration of 100 µg/mL. After incubation for 2 h and 18 h, the cells were washed with Hanks' balanced salt solution (HBSS) thrice and stained with 100 nM LysoTracker Red DND-99 (Thermo Fisher Scientific, Australia) for 5 min. After rinsed with HBSS once, the cells were mounted in 1 mL HBSS and observed under a LSM780 laser scanning confocal microscope (Carl Zeiss). An incubation chamber was equipped on the LSM780 to provide the cells with an environment of 5 % CO2 and 37 °C. The observation used a 100 × oil lense (1.4 N.A.), a Diode 405-30 and an argon lasers. ZEN2012 software (Zeiss) was used for image acquisition and processing.

In order to quantify the cellular uptake, AsPC-1 were incubated with the micelles in 24 well tissue culture plates at a density of 1.5×10^5 cells per well and incubated for 2 days at 37 °C and 5 % CO_2. The micelles were diluted with MilliQ water to 100 µg/mL and sterilised by passing through a sterile 0.45

µm membrane. 500 µL micelles were loaded to each well together with 500 µL 2 × concentrated cell culture medium (3 wells per time point). After incubation for 2 h and 18 h, the cell culture media were collected and freeze-dried. 1 mL dimethylformamide (DMF) was added to the lyophilised powder to dissolve the polymer. The mixture was sonicated for 30 min, shaken for 1h, and filtered through 0.45 µm membranes to remove the precipitations. The fluorescence intensity (FI) was measured with a Cary Eclipse fluorescence spectrophotometer (Agilent). The absorption and emission were 416 and 507 nm, respectively. The reading was zeroed with fresh cell culture medium. The micelles mixed with 2 × medium (1:1) was used as the control. The uptake ratio was calculated with the following equation:

$$\text{Uptake ratio (\%)} = \frac{FI\ (control) - FI(sample)}{FI\ (control)} \times 100\ \%$$

After incubation with micelles for 2 h, AsPC-1 cells took up 10.74 ± 5.96 % of micelles. The micelles were continuously internalizing into cells in an 18 h incubation period. The uptake ratio increased to 18.76 ± 8.30 % after 18 h.

Example 3: Fabrication and characteristics of the amphiphilic block copolymer micelles loaded with a compound

Loaded amphiphilic block copolymer micelles were made. To this end, 2 mg of block copolymer and 1 mL of stock solution (concentration = 1 mg·mL^{-1} in DMF) of the encapsulated material (Nile Red, Curcumin or Doxorubicin) was dissolved in 0.4 mL of DMF. The solution was placed on a stir plate with a high stirring rate at room temperature. Deionized water (3.6 mL) was added dropwise to the solution with a flow rate of 0.2 mL·h^{-1}, leading to a total polymer concentration of 0.4 mg·mL^{-1}. Afterwards the solution was placed in a dialysis membrane with pore size M_w < 3 500 g·mol^{-1} and dialyzed against deionized water for 48 hrs.

Different characteristics of micelles of PPV-*b*-PEGMA, PPV-*b*-PHEA or PPV-*b*-PHPMA and loaded with nile red (NR), curcumin (Cur) or doxorubicin (Dox) are displayed in the table below: intensity (I_{mean}), volume (V_{mean}) and number (N_{mean}) average sizes as obtained through dynamic light scattering (DLS), the average size (D_{50}) as obtained through transmission electron microscopy (TEM), the maximum wavelength (λ_{max}) of the absorption (Abs) and emission (Em) spectrum, the zeta potential as obtained through DLS and the loading efficiency (η).

Loading		**DLS nm**				**TEM nm**	**λ_{max} nm**		**Zeta potential**	**η**
	mg	***I*mean**	***V*mean**	***N*mean**	***Đ***	***D*50**	***Abs***	***Em***	**mV**	**%**
PPV-*b*-PEGMA		119.3	109.2	94.5	0,197	22 ± 2.0	357	466	-28.9	0
NR	0.5	300.7	450.4	116	0.335	n.a.	543	650	-37.3	28.9
Cur	0.5	672.4	812.3	109.4	0.485	30 ± 2.5	428	538	-43.2	27.8
Dox	0.5	260.5	262.2	259.2	0.943	30 ± 2.5	480	593	-35.6	9.1
PPV-*b*-PHEA		183.5	182.5	156.7	0.337	30 ± 1.5	389	491	-25.3	0
NR	0.5	148.5	149.1	147.5	1	n.a.	543	625	-32.9	36.3

Cur	0.5	163.3	163.9	161.4	0.985	n.a.	428	525	-28.0	28.4
Dox	0.5	399.3	404.9	390.7	0.765	n.a.	480	575	-27.9	14.9
PPV-*b*-PHPMA		364.4	341.7	128.3	0.363	50 ± 5	421	495	-38.7	0
NR	0.5	120.5	120.7	119.4	1	n.a.	543	650	-32.8	25.5
Cur	0.5	71.38	71.31	70.26	0.874	n.a.	428	515	-22.4	11.1
Dox	0.5	700.3	714.3	699.4	0.743	n.a.	480	575	-1.58	10.1

We now refer to Fig. 13. The cytotoxicity of the curcumin (a) and doxorubicin (b) loaded PPV-*b*-PEGMA micelles inside living cells was investigated using the protocol described in example 2. Whereas earlier no cytotoxicity was apparent for the unloaded PPV-*b*-PEGMA, a cytotoxicity corresponding to a half maximal inhibitory concentration of 1,18 µM and 1,51 µM was observed for PPV-*b*-PHPMA loaded with curcumin or doxorubicin, respectively. As the unloaded PPV-*b*-PEGMA showed no toxicity, the observed toxicity may thus be attributed to the loaded drugs themselves. As such, this proves that the drugs were successfully loaded into the micelles and subsequently successfully released inside the cells.

The cellular uptake of the AsPC-1 cells, incubated for 72 h, of doxorubicin loaded PPV-*b*-PEGMA micelles was investigated using confocal fluorescence microscopy and differential interference contrast microscopy (DIC); following the protocol described in example 2. The confocal fluorescence of PPV-*b*_PEGMA loaded with doxorubicin, the fluorescence of the cell lysosomes, the DIC of the corresponding cell sample and an overlay of all 3 were imaged. A clear uptake of the loaded micelles in the cell and subsequent release of the payload was nicely confirmed by confocal microscopy results.

It is to be understood that although preferred embodiments, specific constructions and configurations, as well as materials, have been discussed herein for devices according to the present invention, various changes or modifications in form and detail may be made without departing from the scope and technical teachings of this invention. For example, any formulas given above are merely representative of procedures that may be used. Functionality may be added or deleted from the block diagrams and operations may be interchanged among functional blocks. Steps may be added or deleted to methods described within the scope of the present invention.

CLAIMS

1.- Use of a micelle (55) of an amphiphilic block copolymer (51) to modify a cell, wherein the amphiphilic block copolymer (51) comprises one or more hydrophobic blocks (52) and one or more hydrophilic blocks (53), wherein at least one block (52, 53) comprises a conjugated polymer, and wherein the micelle (55) is optionally loaded with a compound (54) therein.

2.- The use according to claim 1, wherein modifying the cell comprises dyeing at least part of the cell and/or comprises releasing the compound (54), if present, inside the cell.

3.- A micelle (55) of an amphiphilic block copolymer (52) usable according to claim 1 or 2, the amphiphilic block copolymer (52) comprising:

i. a first block (52) comprising a poly(*p*-arylene vinylene) and

ii. a non-ionic second block (53) comprising a vinyl polymer;

wherein at least one block (52, 53) is hydrophobic and at least one other block (53, 52) is hydrophilic.

4.- The micelle (55) according to claim 3, being present in an aqueous medium.

5.- The micelle (55) according to any of claims 3 or 4, wherein the poly(*p*-arylene vinylene) is selected in such a way that it is luminescent in solution when in a non-micellar form.

6.- The micelle (55) according to any of claims 3 to 5, being loaded with a compound (54) therein.

7.- The micelle (55) according to any of claims 3 to 6, wherein the compound (54) is a contrast agent, a drug or a polynucleotide.

8.- A method for forming the micelle (55) of any of claims 3 to 7, comprising the steps of:

a. dissolving the amphiphilic block copolymer (51) in a first liquid and

b. mixing thereto a second liquid;

wherein either the first or second liquid is an aqueous liquid and wherein the other liquid is a non-aqueous liquid.

9.- The method according to claim 8, wherein the first liquid is dimethylformamide and wherein the second liquid is water.

10.- The method according to claim 8 or 9, comprising an additional step of:

c. removing the first liquid.

11.- The method according to any of claims 8 to 10, wherein an additional compound (54) is present in the first and/or the second liquid, thereby forming the micelle (55) loaded with the compound (54) therein.

12.- An amphiphilic block copolymer (51) usable to make the micelle (55) of any of claims 3 to 7, comprising:

i. a first block (52) comprising a poly(*p*-arylene vinylene) and

ii. a non-ionic second block (53) comprising a vinyl polymer (53);

wherein at least one block (52, 53) is hydrophobic and at least one other block (53, 52) is hydrophilic.

13.- The amphiphilic block copolymer (51) according to claim 12, where the block copolymer (51) is a di- or tri-block copolymer (51).

14.- The amphiphilic block copolymer (51) according to claim 12 or 13, wherein the poly(*p*-arylene vinylene) comprises hydrophobic substituents.

15.- The amphiphilic block copolymer (51) according to any of claims 12 to 14, wherein the vinyl polymer (53) is obtainable by the polymerization of a monomer obtainable from the reaction of (meth)acrylic acid with respectively an alcohol of general formula HO-R or an amine of general formula NH_2-R, wherein R is non-ionic.

16.- The amphiphilic block copolymer (51) according to any of claims 12 to 15, wherein the vinyl polymer (51) is a hydrophilic poly(ethylene glycol monomethyl ether methacrylate), poly(2-hydroxyethyl acrylate), or poly(n-(2-hydroxypropyl) methacrylamide), or a polystyrene.

17.- The amphiphilic block copolymer (51) according to any of claims 12 to 16, wherein the poly(*p*-arylene vinylene) is a poly(2-methoxy-5-(3,7-dimethyloctyloxy)-1,4-phenylene vinylene) or poly(2-methoxy-5-(2-ethylhexyloxy)-1,4-phenylenevinylene).

18.- The amphiphilic block copolymer (51) according to any of claims 12 to 17, wherein the conjugated polymer (52) has a length of 7 repeating units or more.

19.- The amphiphilic block copolymer (51) according to any of claims 11 to 18, wherein the copolymer is a poly(*p*-arylene vinylene)-*block*-poly(meth)acrylate ester, a poly(*p*-arylene vinylene)-*block*-poly(meth)acrylamide or a poly(*p*-arylene vinylene)-*block*-polystyrene.

20.- A method for synthesizing an amphiphilic block copolymer (51) in accordance with any of claims 11 to 19, comprising the steps of:

a. providing an initiator comprising a first initiating moiety for an anionic polymerization and a second initiating moiety for a living radical polymerization,

b. polymerizing, in an anionic initiation pathway, a poly(*p*-arylene vinylene) monomer with the first initiating moiety, so as to form a precursor to a poly(*p*-arylene vinylene) first block,

c. polymerizing, in a living radical polymerization pathway, a vinyl monomer with the second initiating moiety, so as to form a second block, and

d. performing a thermal elimination, so as to obtain the poly(*p*-arylene vinylene) first block.

21.- The method according to claim 20, wherein the initiator is 4-((methylsulfinyl)methyl)phenyl-2-bromo-2-methylpropanoate.

22.- The method according to claim 20 or 21, wherein the poly(*p*-arylene vinylene) monomer is a 1-(chloromethyl)-5-alkoxy-2-alkoxy-4-((octylsulfinyl)methyl)benzene.

23.- The method according to any of claims 20 to 22, wherein the vinyl monomer is a hydrophilic 2-hydroxyethyl acrylate, ethylene glycol monomethyl ether methacrylate, or n-(2-hydroxypropyl) methacrylamide, or a hydrophobic polystyrene.

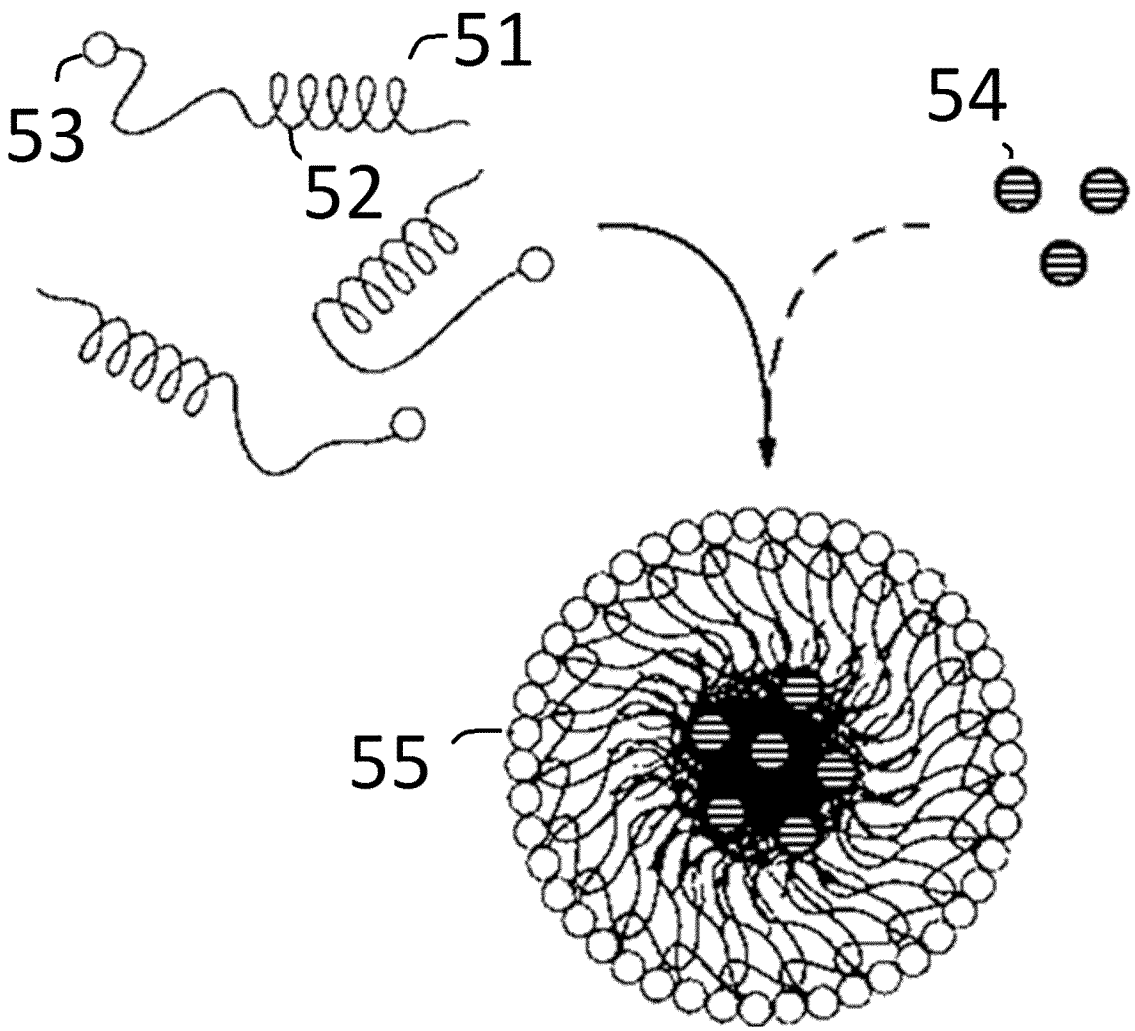


FIG. 1

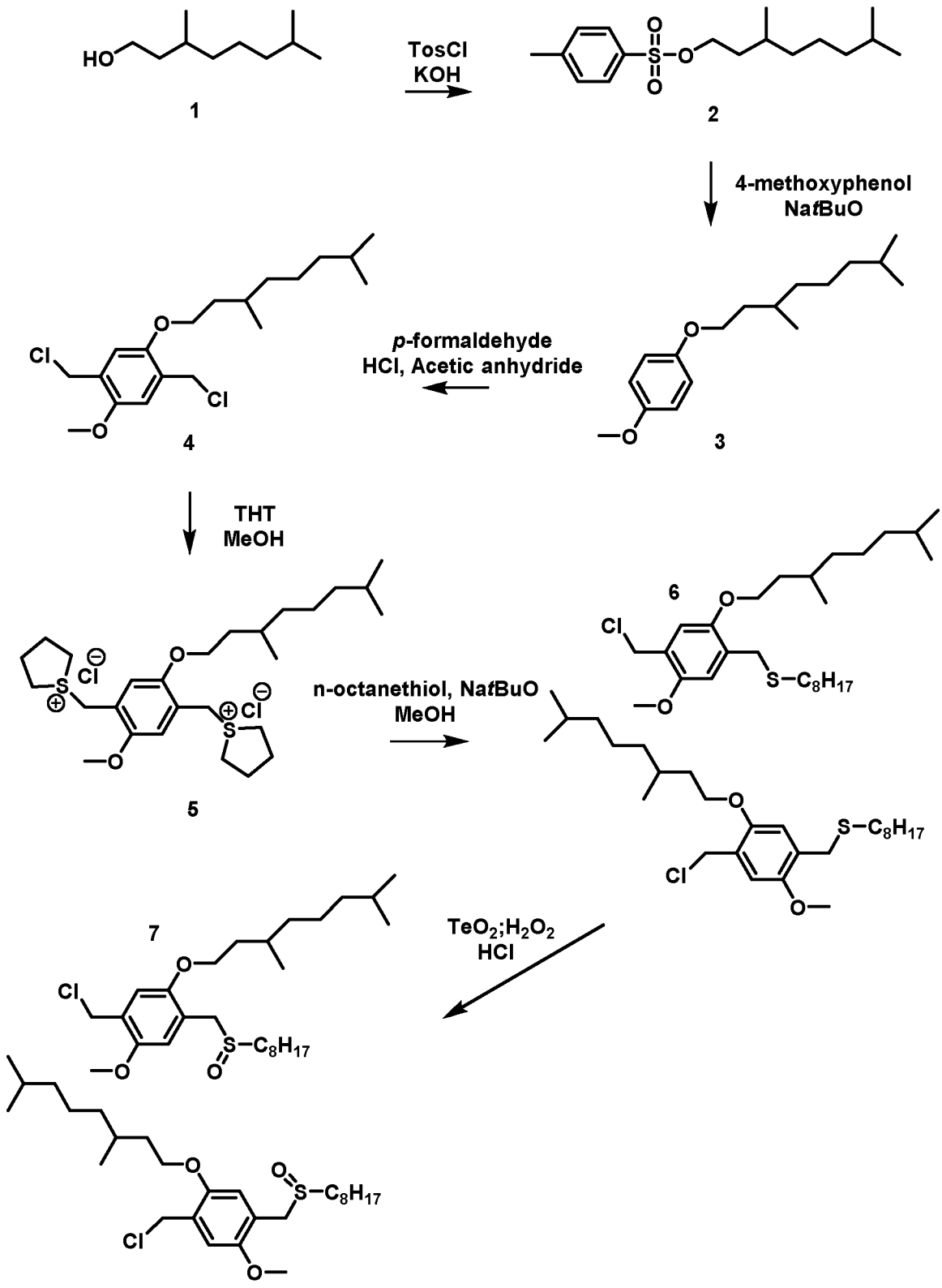

HO
1
TosCl
KOH
2
4-methoxyphenol
NatBuO
3
p-formaldehyde
HCl, Acetic anhydride
4
THT
MeOH
5
n-octanethiol, NatBuO
MeOH
6
TeO2;H2O2
HCl
7

FIG. 2

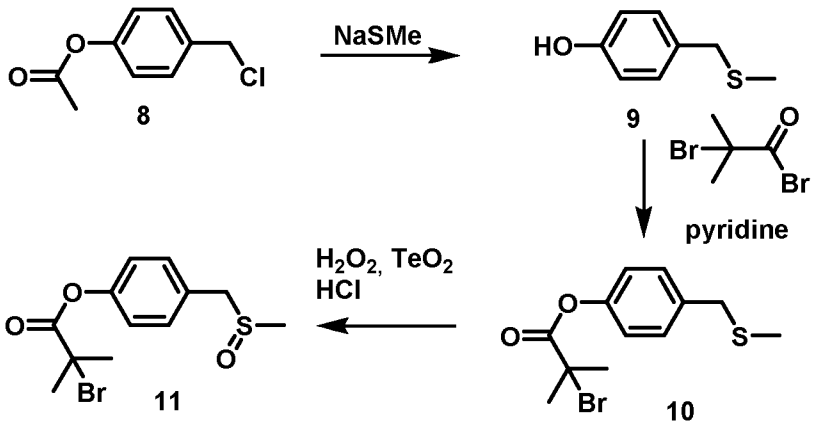

NaSMe
8
9
pyridine
H_2O_2, TeO_2
HCl
10
11

FIG. 3

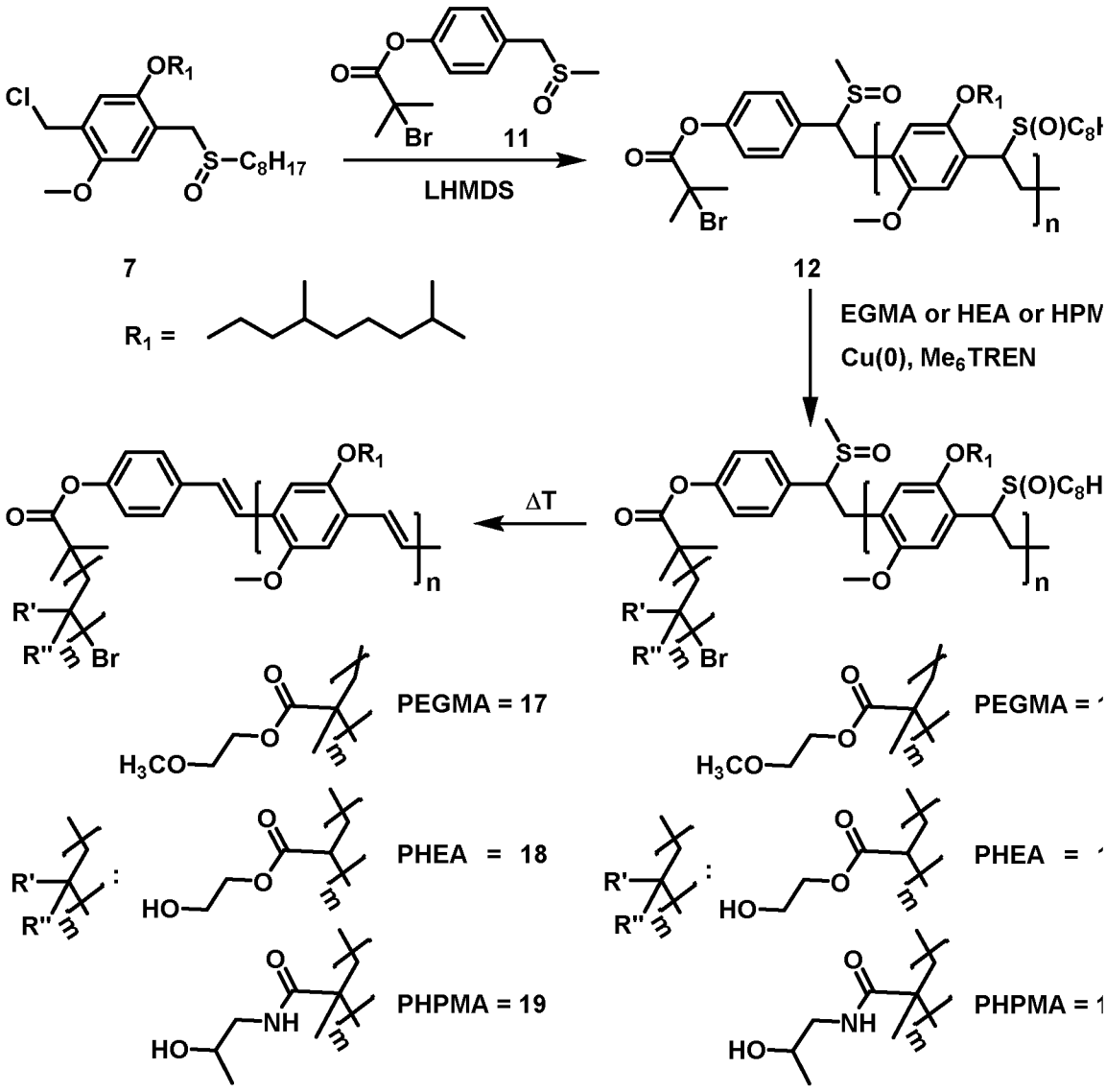

LHMDS
11
7
12
R_1 =
EGMA or HEA or HPM
Cu(0), Me_6TREN
ΔT
PEGMA = 17
PHEA = 18
PHPMA = 19

FIG. 4

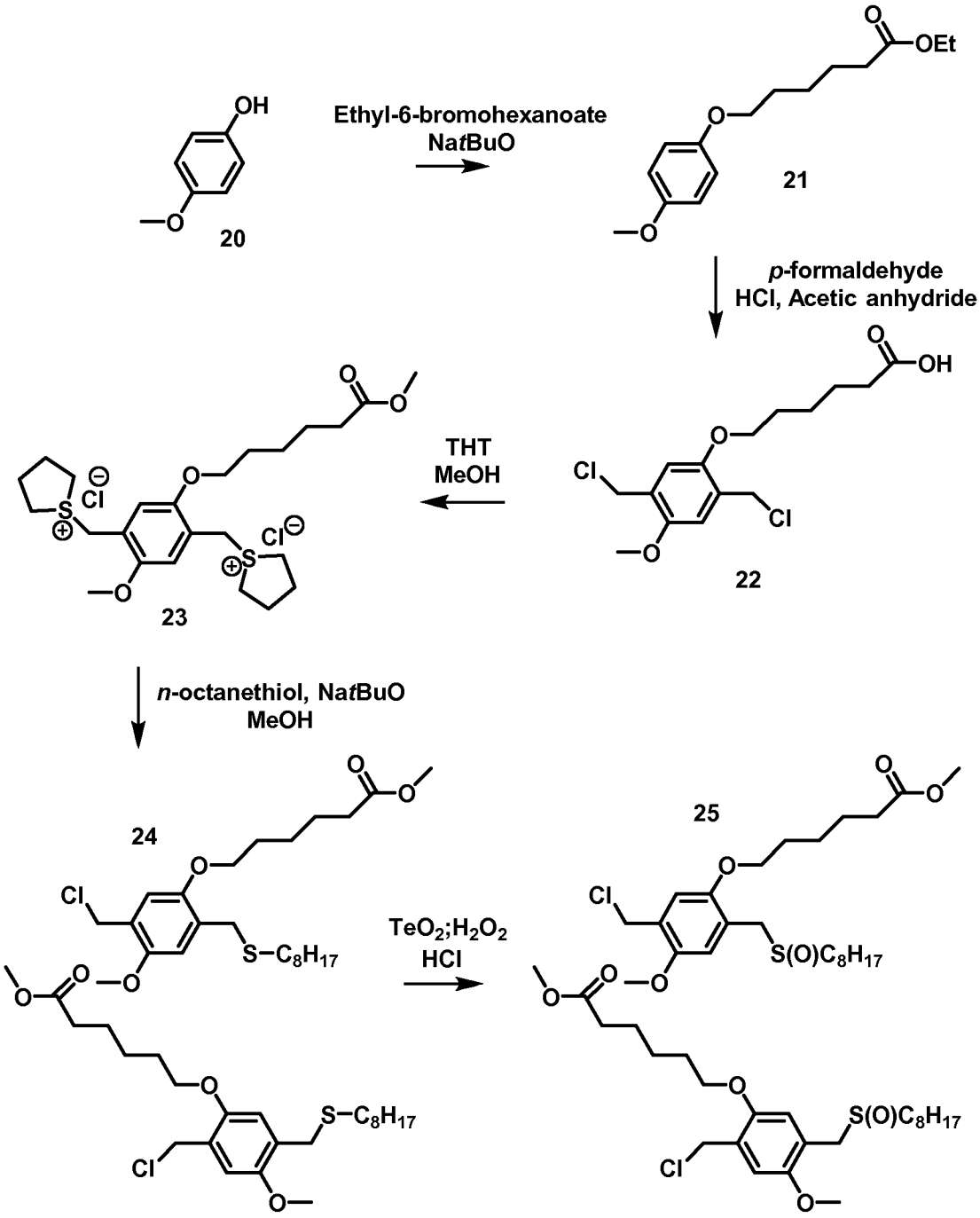

OH
20
Ethyl-6-bromohexanoate
NatBuO
O
OEt
21
p-formaldehyde
HCl, Acetic anhydride
OH
Cl
22
THT
MeOH
23
n-octanethiol, NatBuO
MeOH
24
S–C8H17
TeO2;H2O2
HCl
25
S(O)C8H17

FIG. 5

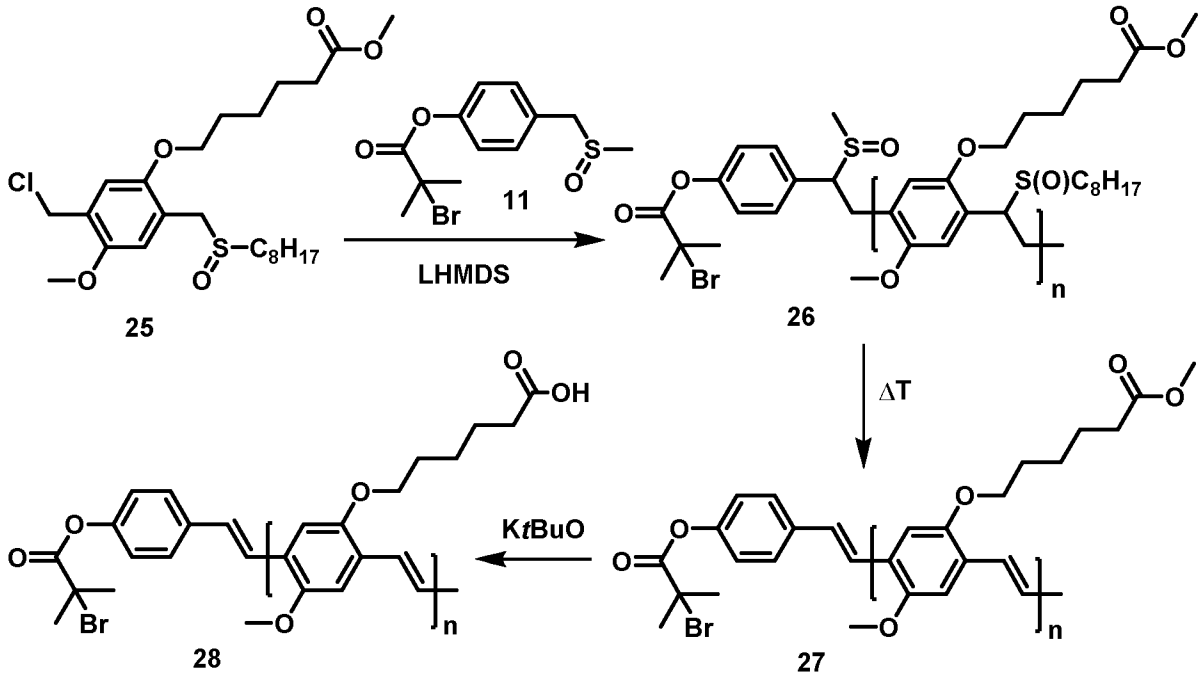

Cl
S–C8H17
11
Br
LHMDS
S(O)C8H17
25
26
ΔT
OH
KtBuO
28
27

FIG. 6

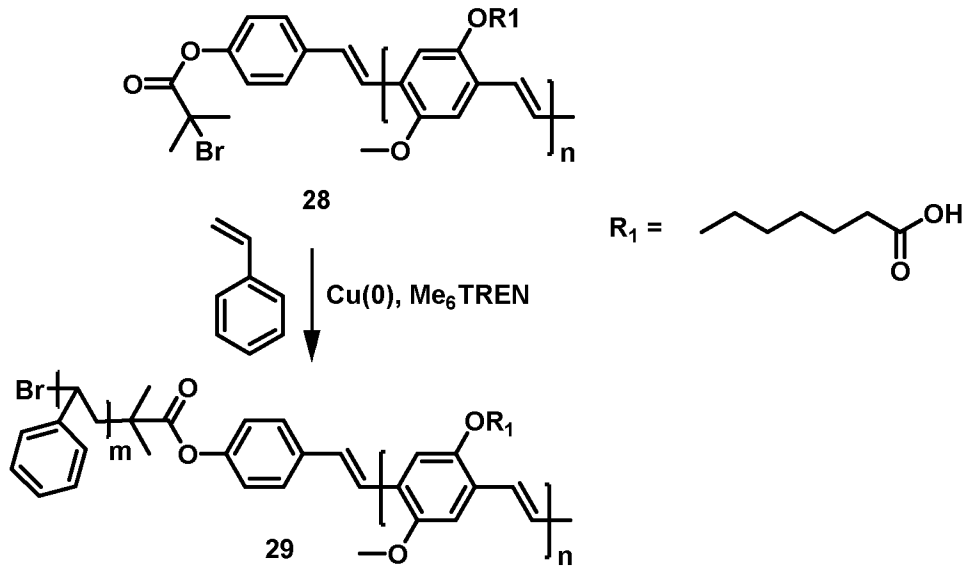

OR1
Br
28
R1 =
OH
Cu(0), Me6TREN
Br
m
OR1
29

FIG. 7

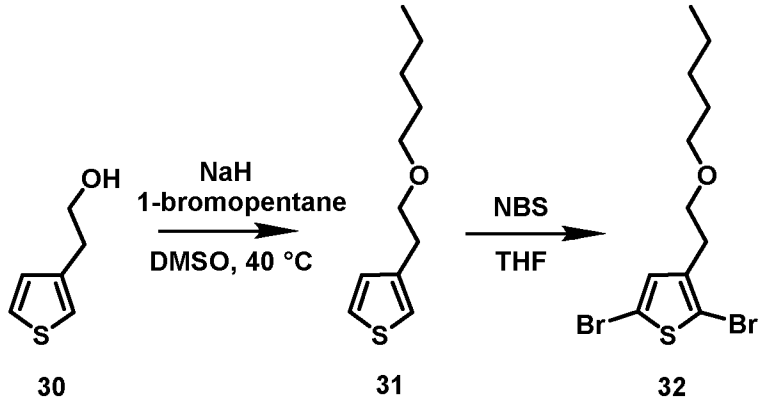


FIG. 8

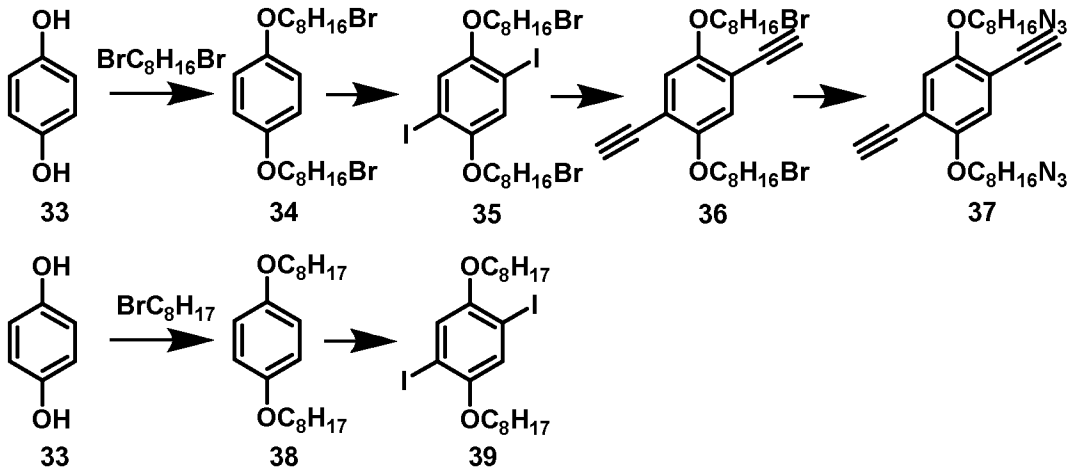


FIG. 9

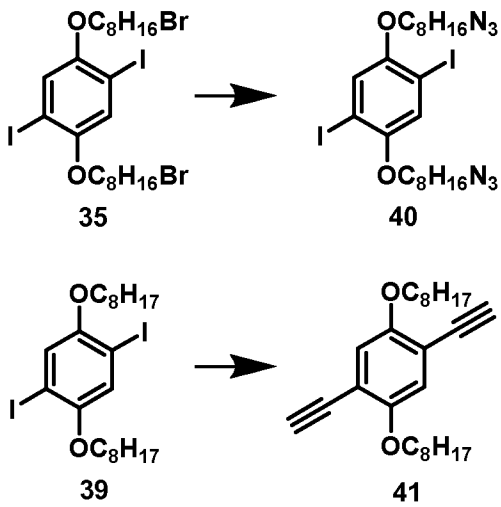


FIG. 10

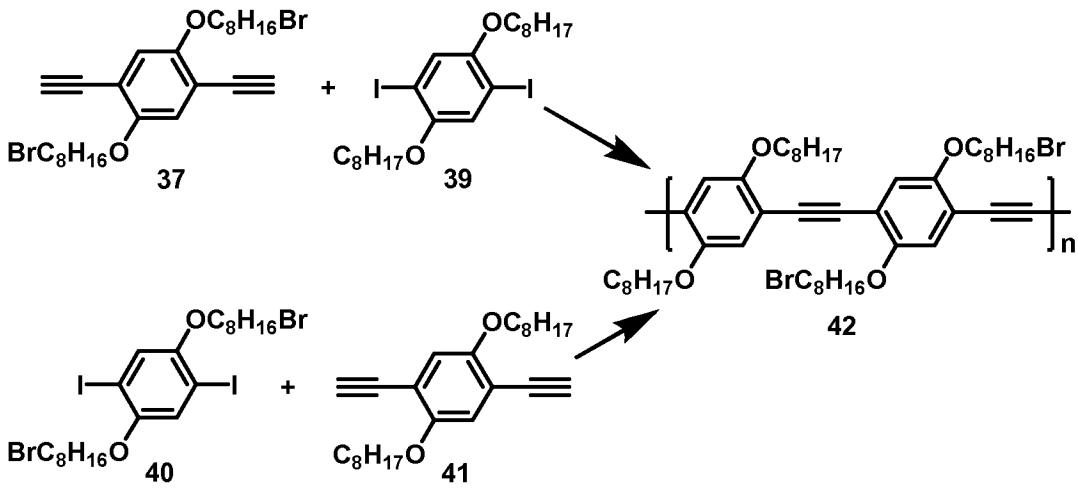

OC8H16Br
BrC8H16O
37
OC8H17
I
I
C8H17O
39
OC8H17
OC8H16Br
C8H17O
BrC8H16O
n
42
OC8H16Br
I
I
BrC8H16O
40
OC8H17
C8H17O
41

FIG. 11

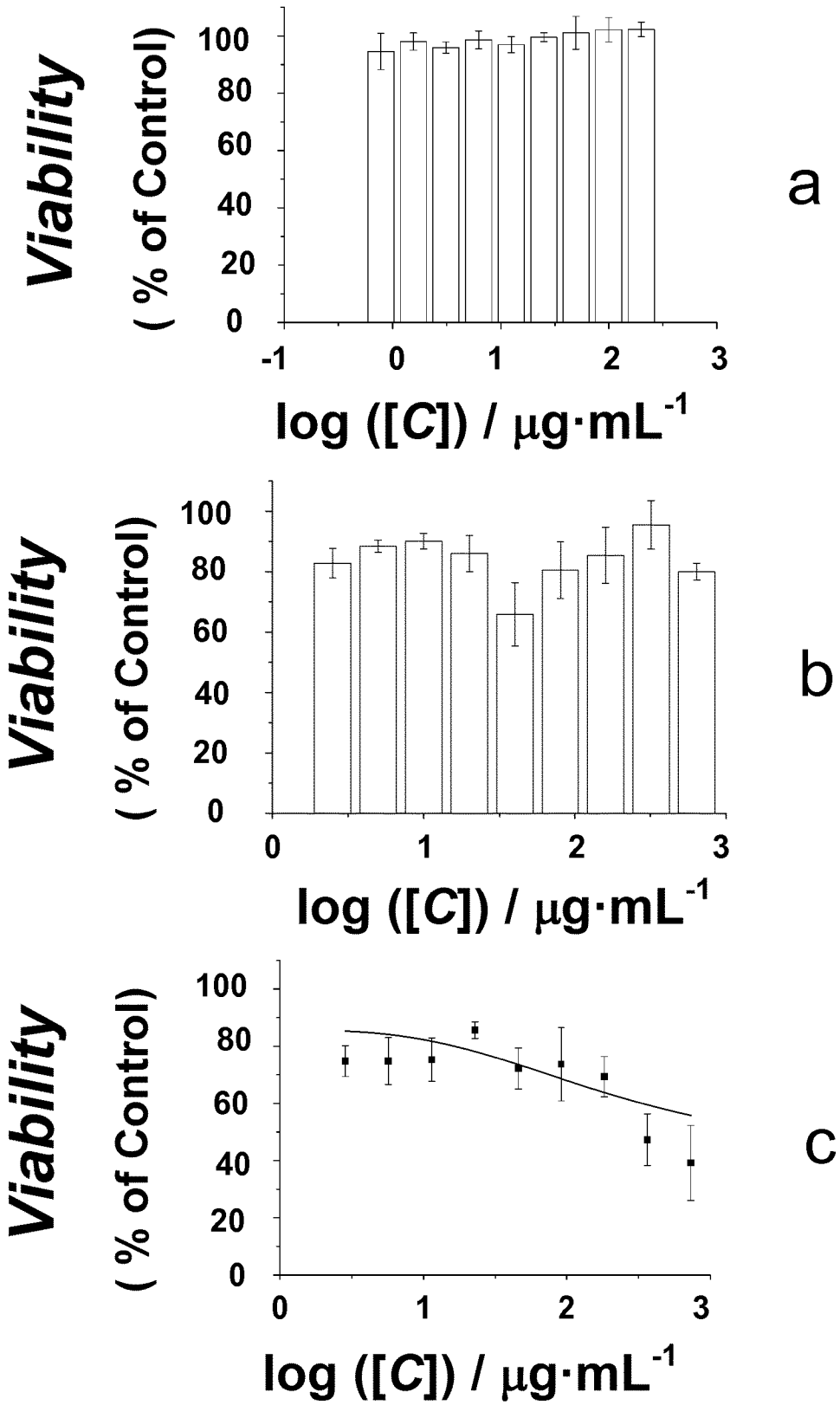
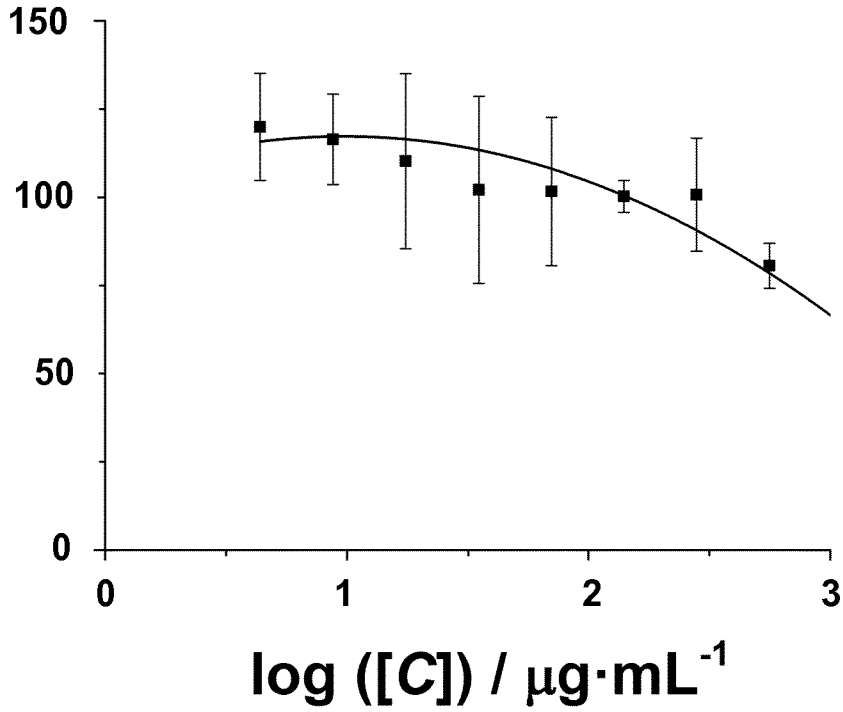
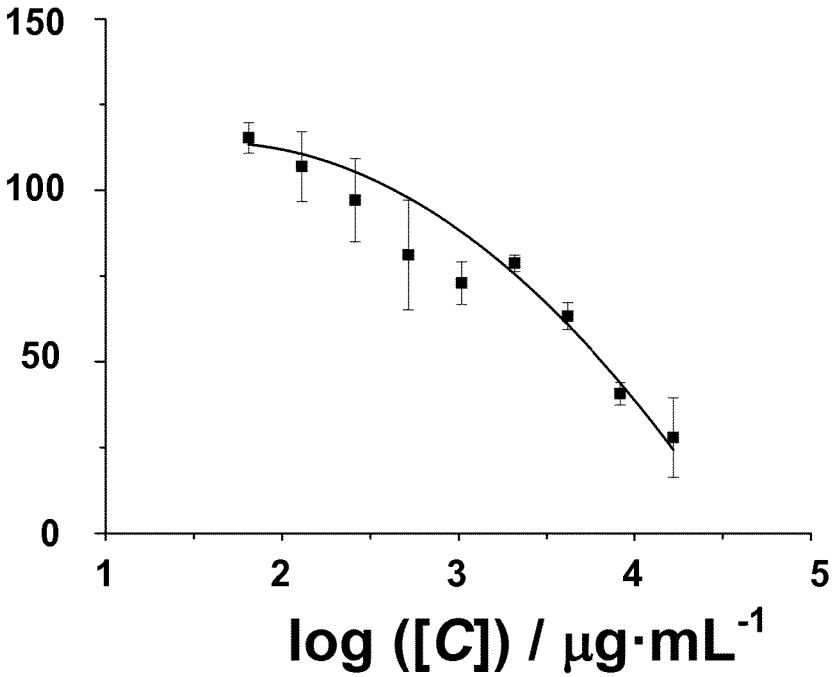

Viability
(% of Control)
100
80
60
40
20
0
-1
0
1
2
3
log ([C]) / μg·mL-1
a
Viability
(% of Control)
100
80
60
40
20
0
0
1
2
3
log ([C]) / μg·mL-1
b
Viability
(% of Control)
100
80
60
40
20
0
0
1
2
3
log ([C]) / μg·mL-1
c

FIG. 12



a



b

FIG. 13

INTERNATIONAL SEARCH REPORT

International application No
PCT/EP2017/059099

A. CLASSIFICATION OF SUBJECT MATTER
INV. A61K9/107 A61K47/32 C08F293/00 C08G61/02
ADD.

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
A61K C08G

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
EPO-Internal, WPI Data, EMBASE, BIOSIS, CHEM ABS Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	INGE COSEMANS ET AL: "Synthesis of well-defined PPV containing block polymers with precise endgroup control by a dual-initiator strategy", POLYMER CHEMISTRY, vol. 4, no. 12, 1 January 2013 (2013-01-01), pages 3471-3479, XP055383399, GB ISSN: 1759-9954, DOI: 10.1039/c3py00303e	3-5, 7-10, 12-14, 17,18, 20-23
Y	abstract page 3471, left-hand column, paragraph 1 - page 3472, right-hand column, paragraph 2 page 3473, right-hand column, paragraph 2 - page 3474, left-hand column, paragraph 3 page 3476, right-hand column, last paragraph - page 3477, left-hand column, paragraph 2 Scheme 5;	6,11

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

[X] See patent family annex.

* Special categories of cited documents :

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

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"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
26 June 2017	30/06/2017

Name and mailing address of the ISA/	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016	Gómez Gallardo, S

1

Form PCT/ISA/210 (second sheet) (April 2005)

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
	page 3477 page 3478, left-hand column, last paragraph - page 3479, left-hand column, paragraph 3	

X	US 2009/281242 A1 (LANDIS STEFAN [FR]) 12 November 2009 (2009-11-12) page 2, paragraph 49 - page 3, paragraph 51 claims 1-8	3,5,7, 12-14, 16,19

X	CHUN-CHIH HO ET AL: "Synthesis and Self-Assembly of Poly(diethylhexyloxy- p -phenylenevinylene)- b -poly(methyl methacrylate) Rod-Coil Block Copolymers", MACROMOLECULES, vol. 42, no. 12, 23 June 2009 (2009-06-23), pages 4208-4219, XP055383385, US ISSN: 0024-9297, DOI: 10.1021/ma802551v the whole document	12-15, 18,19

X	COSEMANS INGE ET AL: "Synthesis of PPV-b-PEG block copolymers via CuAAC conjugation", EUROPEAN POLYMER JOURNAL, vol. 55, 28 March 2014 (2014-03-28), pages 114-122, XP028652953, ISSN: 0014-3057, DOI: 10.1016/J.EURPOLYMJ.2014.03.016 abstract page 114, left-hand column, paragraph 1 - page 116, left-hand column, paragraph 1 page 120, right-hand column, last paragraph - page 122, left-hand column, paragraph 1	1,2

Y	WO 2013/055791 A1 (UNIV MICHIGAN [US]) 18 April 2013 (2013-04-18) page 2, paragraph 5 - page 3, paragraph 7 page 12, paragraph 65 - page 14, paragraph 70 page 24, paragraph 100 - page 25, paragraph 101	6,11

Y	US 2013/078310 A1 (SILL KEVIN [US] ET AL) 28 March 2013 (2013-03-28) page 1, paragraph 7-11 page 2, paragraphs 19,20	6,11

1

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No
PCT/EP2017/059099

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
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		FR 2930244 A1	23-10-2009
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