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The synthesis and characterisation of masked phosphonioalkyl selenoates: Potential ligands for the production of functionalised gold nanoparticles

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Abstract

Two new masked phosphonioalkylselenoate ligands, bis(3-triphenylphosphoniopropyl)diselenide- and 6-(selenocyano)hexyl-triphenylphosphonium-selenocyanates, have been prepared. The molecular structure of the bis(3-triphenylphosphoniopropyl)diselenide diselenocyanate has been determined by X-ray crystallography. The structure reveals an overall stoichiometry of $\{[Ph_3P^+(CH_2)_3Se]_2(SeCN^-)_2 \cdot KOH\}$, with the bis(3-triphenylphosphoniopropylselenium)diselenocyanate units arranged in pairs around an inversion centre. The potassium ion is disordered over several positions but its main component forms a near linear K···Se contact to one of the selenium atoms in the diselenide bond. The hexyl derivative, 6-(selenocyano)hexyl-triphenylphosphonium selenocyanate forms as a yellow oil that was characterised spectroscopically. Both phosphonioalkylselenide cations undergo reductive cleavage to form phosphonioalkylselenoate zwitterions. Attempts to prepare phosphonioalkylselenoate-functionalised gold nanoparticles *in situ* through the $NaBH_4$-promoted reduction of tetrachloroaurate salts in a water/dichloromethane biphasic system are also described.

Keywords: Phosphonium salt; Selenium; Nanoparticle; Gold; Crystal structure

1. Introduction

There is considerable current interest in the synthesis of monolayer-protected gold nanoparticles (MPCs) [1,2], particularly receptor-functionalised species which are able to recognize and interact with specific biomolecules, as part of the search for new diagnostic biosensor technologies and novel therapeutic agents [3]. The most widely studied surface-groups are organic thiolates (RS^-), usually derived from the corresponding thiols through the *in situ* reduction of a gold salt in the presence of the parent ligand.

We have recently reported the synthesis of a new family of phosphonioalkyl thiosulfate zwitterions which behave as masked thiolate ligands producing cationic-functionalised MPCs, which have potential as diagnostic biorecognition systems [4]. The term 'masked thiolate' is used here to describe alkane thiolate species which are 'protected' as another group, in this case thiosulfate, and which, upon contact with metal surfaces, undergo cleavage of the sulfur–sulfur bond generating the thiolate anion which attaches to the metal surface.

Phosphonium species are well suited for the production of biocompatible MPCs and are being investigated as agents for targeted dug delivery to cell mitochondria [5–8]. However, although it is well established that tertiary phosphines and phosphine oxides, such as TOPO [9], are effective passivating ligands for the production of MPCs, other phosphorus-containing species have received comparatively little attention. The few available reports include

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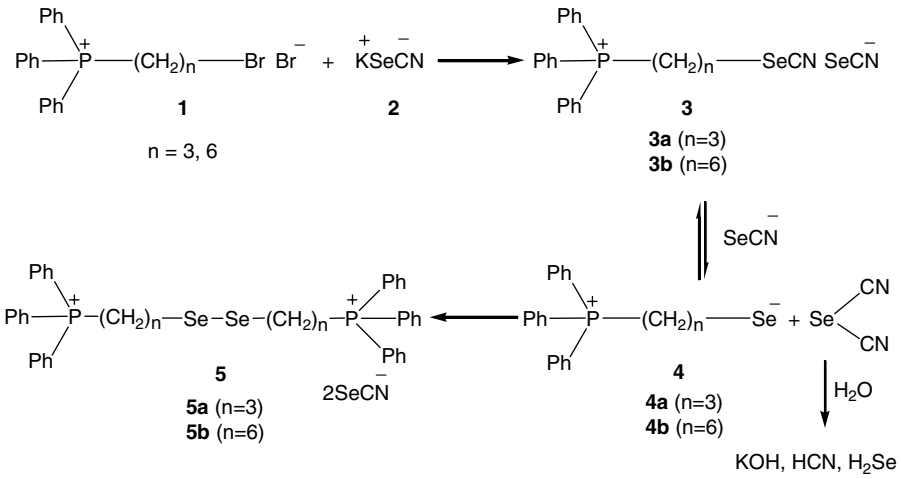
studies of CdSe nanoparticles capped with the phosphonium ionic liquid trihexyl(tetradecyl)phosphonium bis(2,4,4-trimethylpentylphosphinate) [10], and gold nanoparticles capped with phosphides (PR_2^-) [11].

In order to extend the range of cationic phosphonium ligands we have prepared two new potential phosphonioalkylselenoate precursor ligands. Most of the compounds used as protecting ligands in the synthesis of functionalised gold nanoparticles reported in the literature are those containing thiol groups. Selenoate anions offer an alternative to thiolates as capping agents for gold surfaces as a result of the similarity of the sulfur and selenium atoms in terms of chemical properties. Despite this similarity, organo-selenols and -diselenides have not received the same attention as the sulfur analogues in this regard and there are only a few examples of organoselenium-stabilised MPCs, although organoselenium species including dialkyl diselenides and dialkyl selenides are known to form self-assembled monolayers (SAMs) on planar noble metal surfaces [12]. For example, Kim and co-workers investigated the adsorption behaviour of benzeneselenol (BSe) and diphenyl diselenide (DPDSe) on a gold substrate by surface enhanced Raman spectroscopy (SERS). They demonstrated that the selenol chemisorbs on gold as the selenolate, and in the case of diselenides, the Se–Se bond is cleaved when the diselenide is in contact with the gold surface, resulting in the formation of a stable monolayer [13]. Zharnikov and co-workers confirmed Se–Se bond cleavage in bis(biphenyl)diselenide on gold and silver substrates by high resolution X-ray photoelectron spectroscopy (HRXPS) [14]. Selenoate-stabilised MPCs have been reported by Ulman and co-workers, who investigated the use of the alkaneselenols and dialkyldiselenides to protect the nanoparticle surface. They developed a one-phase preparation of alkaneselenol-protected gold nanoparticles and also provided a detailed study of the gold nanoparticle morphology, structure and bonding preference as compared to their alkanethiolate analogues. As is well known, when the S/Au mole ratio decreases from 2:1 to 1:1 to 1:2 to 1:3 in the synthesis of alkanethiol-protected gold nanoparticles, the average particle size increases. However, in the case of the dialkyldiselenide/Au ratio, no such tendency and no significant dependence were observed [15]. Tong and co-workers synthesized octaneselenol-protected gold nanoparticles using the method developed by Brust and Schiffrin, with dioctyldiselenide as the precursor of the protecting ligand. They were the first to report the observation of ^{77}Se NMR signals from the octaneselenoate-protected gold nanoparticles, showing the potential of NMR as a powerful tool to investigate the interactions of the ligand with the nanoparticle surface [16].

In this paper we report the synthesis and characterisation of bis(3-triphenylphosphoniopropyl)diselenide- and 6-(selenocyano)hexyltriphenylphosphonium-selenocyanates which have the potential to act as 'masked selenoate' ligands for the preparation of gold MPCs. Our studies into the reaction of these ligands with gold nanoparticles formed *in situ* through the $NaBH_4$-promoted reduction of tetrachloroaurate salts are also described.

2. Results and discussion

Our synthetic strategy is outlined in Scheme 1 and is based on the protocol we have reported previously for the preparation of phosphonioalkyl 'masked' thiolate ligands [4]. Initially the synthesis of the selenocyanopropyl phosphonium salt (**3a**, $n = 3$) was planned as a possible source of the phosphonioalkylselenide zwitterion (**4**) after the reduction of (**3**) with $NaBH_4$ or Au^0 during gold nanoparticle functionalisation. However, spectroscopic data indicated that the bis(3-triphenylphosphoniopropyl)diselenide–di(selenocyanate) salt (**5a**, $n = 3$), rather than **3a**, was obtained by treatment of the bromopropylphosphonium salt (**1a**, $n = 3$) with an excess of potassium selenocyanate in aqueous ethanol. It is likely that the diselenide (**5a**) is formed by *in situ* oxidation of the phosphonioselenoate



Scheme 1. Synthesis of bis(3-triphenylphosphoniopropylselenium)diselenocyanate (**5a**) and 6-(selenocyano)hexyl-triphenylphosphonium selenocyanate (**3b**).

(**4a**, $n = 3$), which may arise as shown. Microanalytical data supported the formulation of **5a** but indicated the presence of 1 mol of KOH per mole of diselenide salt. Electrospray mass spectrometry also supported the formulation of this compound as containing the bis(3-phosphoniopropyl)diselenide cation. When studied by ESMS in positive ion mode, ions corresponding to 1/2M+H, 1/2M+Na, 1/2M+Se and M+K were observed.

Table 1
Selected bond lengths (Å) and angles (°) in bis(3-triphenylphosphoniopropyl)diselenide–di(selenocyanate) (**5a**)

P1–C19	1.79(6)	Se1–C21	1.96(7)
P1–C7	1.79(6)	Se1–Se2	2.318(10)
P1–C1	1.79(6)	Se2–C22	1.97(6)
P1–C13	1.80(6)	Se3–C43	1.74(12)
P2–C37	1.79(6)	C43–N1	1.18(11)
P2–C24	1.79(6)	Se4–C44	1.83(9)
P2–C25	1.79(6)	C44–N2	1.12(9)
P2–C31	1.80(6)		
C19–P1–C7	111(3)	C8–C7–P1	119(5)
C19–P1–C1	109(3)	C12–C7–P1	122(5)
C7–P1–C1	109(3)	C18–C13–P1	121(5)
C19–P1–C13	110(3)	C14–C13–P1	118(5)
C7–P1–C13	108(3)	C20–C19–P1	114(4)
C1–P1–C13	110(3)	C20–C21–Se1	114(5)
C37–P2–C24	112(3)	C23–C22–Se2	114(4)
C37–P2–C25	108(3)	C23–C24–P2	114(4)
C24–P2–C25	109(3)	C30–C25–P2	120(5)
C37–P2–C31	108(3)	C26–C25–P2	120(5)
C24–P2–C31	109(3)	C32–C31–P2	118(5)
C25–P2–C31	110(3)	C36–C31–P2	121(5)
C21–Se1–Se2	100(2)	C38–C37–P2	120(5)
C22–Se2–Se1	100.4(19)	C42–C37–P2	121(5)
C6–C1–P1	120(5)	N1–C43–Se3	179(8)
C2–C1–P1	120(5)	N2–C44–Se4	178(7)

Salt **5a** was re-crystallised from DCM–ether as a yellow crystalline solid suitable for single crystal X-ray diffraction. X-ray analysis confirmed the formulation of **5a** as a diselenide rather than as the selenocyanate (**3a**) originally anticipated. Selected bond lengths [Å] and bond angles [°] in **5a** are presented in Table 1. The molecular structure of **5a** (Fig. 1) contains Se–Se and C–Se–Se bonds which indicate the presence of a dialkyldiselenide group in the molecular structure. The Se–Se bond length in **5a** [2.318(10) Å] is similar to those found in other diorganodiselenides (R–Se–Se–R). Although the molecular structures of a number of diorgandiselenides have been reported, the majority contain aryl groups or sterically demanding alkyl groups as substituents [17]. Previous studies of diorganodiselenides have shown that the Se–Se and Se–C bond lengths are largely independent of the organic substituents and steric strain is relieved by increasing the C–Se–Se–C dihedral angle [17,18]. The phosphonioalkyl groups in **5a** are not particularly sterically demanding and the dihedral angle C(22)–Se(2)–Se(1)–C(21) [62.80(4)°] is significantly lower than those with bulkier groups, which typically lie in the range 73-104° [17]. The bond lengths and angles around the triphenylphosphonium moieties are similar to those in the comparable 3-triphenylphosphoniopropylthiosulfate zwitterion [4]. The crystal structure of **5a** shows two N–C–Se bond systems, indicating the presence of two selenocyanates as counterions (two anions per molecule for neutrality), and confirms the presence of K^+ and OH^- ions in the unit cell. The overall structure consists of bis(3-triphenylphosphoniopropylselenium)diselenocyanate units arranged in pairs around an inversion centre. The potassium ion is disordered over several positions but its main component forms a near linear [172.99(7)°] K···Se contact of

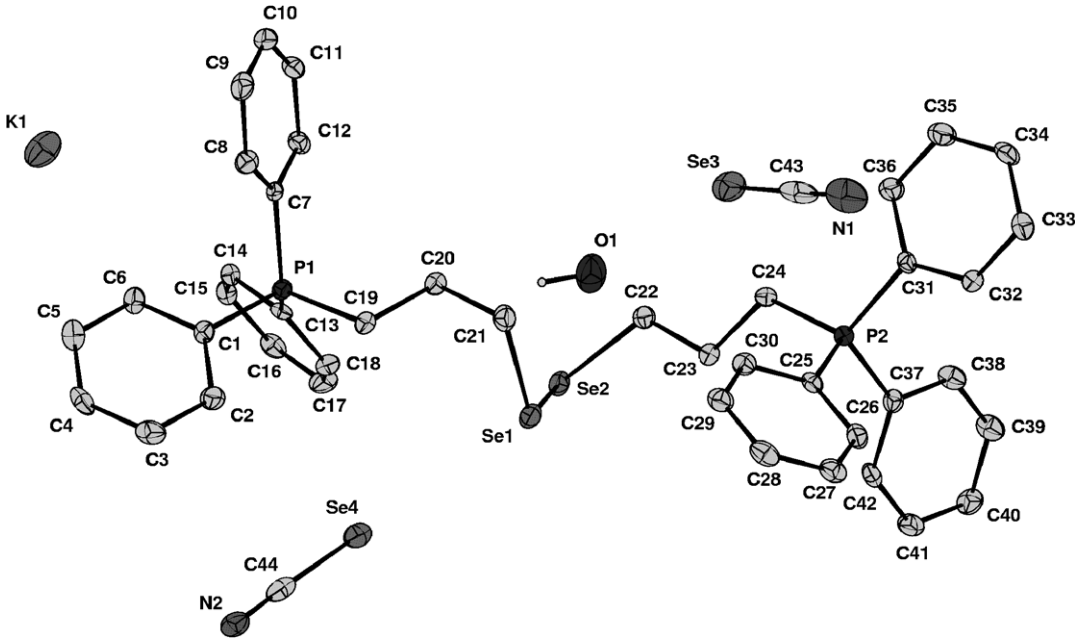


Fig. 1. Molecular structure of $\{[Ph_3P^+(CH_2)_3Se]_2(SeCN^-)_2 \cdot KOH\}$ (**5a**).

3.459(6) Å to the diselenide group, which is similar to the K···Se distances found in other compounds [19]. Thus, for example, potassium 2-methoxybenzenecarboselonate has K–Se bond lengths in the range 3.309(1)–3.625(2) Å [20]. One of the two independent selenocyanate anions forms a slightly bent hydrogen bond (O1W–H1W···N2 = 166°) to the OH anion; the other is involved in Se···K interactions to the disordered potassium (the exact nature of these interactions is hard to assess due to the disorder). Fig. 2 shows interactions between the K and the Se atoms corresponding to the diselenide group of the molecule and Se atom of the counterion. These K···Se interactions [and the O–H···N hydrogen bonds (Table 2)] influence the packing of the cations and anions in the unit cell.

Since the synthesis of the propyl analogue did not follow the expected pathway the synthesis of the six carbon chain analogue of **5a** was attempted by following the same synthetic route. However, 6-(selenocyano)hexyl-triphenylphosphonium selenocyanate (**3b**, $n = 6$), rather than **5b** (**5**, $n = 6$), was obtained. NMR spectroscopy supported the formulation of this compound. Compound **3b** forms as a yellow oil. The longer chain 3-triphenylphosphonioalkylthiosulfates also tend to form as oils or waxy solids [4]. When studied by MALDI TOFMS in positive ion mode (accurate mass analysis), an ion corresponding to M+H was observed. It may be that the longer carbon chain

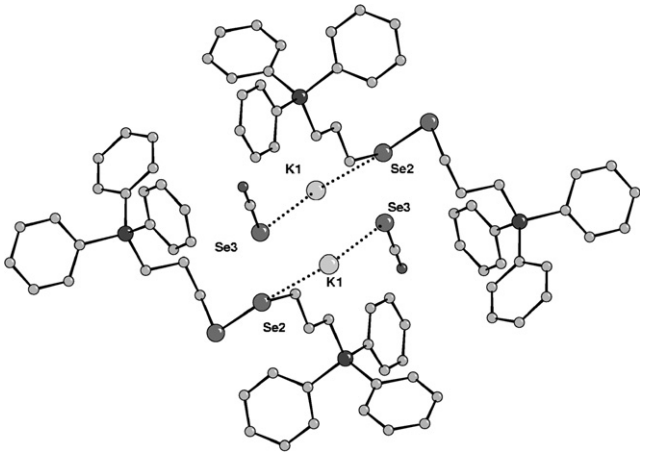


Fig. 2. Intramolecular contacts showing possible K···Se interactions between the K cations and the Se atoms of the diselenide groups and the corresponding to the selenocyanate counterions in bis(3-triphenylphosphoniopropyl)diselenide–di(selenocyanate) (**5a**).

Table 2
Intramolecular contacts for hydrogen bonding between O–H of the KOH moiety and nitrogen atom of the selenocyanate counterion in bis(3-triphenylphosphoniopropyl)diselenide–di(selenocyanate) (**5a**)

D–H···A	*d*(D–H)	*d*(H···A)	*d*(D···A)	∠(DHA)
O1W–H1W···N2^{i}	0.900(16)	2.1(3)	3.03(9)	166(100)

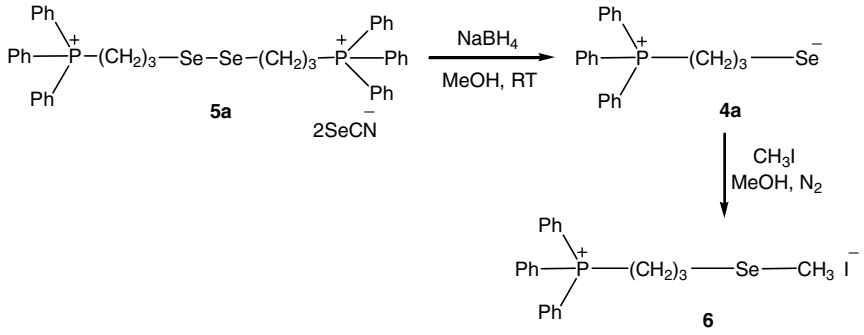
Symmetry transformations used to generate equivalent atoms: (i) $-x+2$, $-y+2$, $-z+1$.

suppresses the inductive effect of the phosphonium group on the cyano group of the alkyl selenocyanato group, such that nucleophilic attack by excess of selenocyanate anions is not favoured.

Previous studies have shown that the Se–Se and Se–OH bonds present in organic diselenides and alkylselenols can be cleaved in the presence of a metal surface and gold nanoparticles, enabling the formation of SAMs and functionalised nanoparticles, respectively. In order to study the capacity of compound **5a** as a protecting ligand and to prove the formation of the zwitterion $Ph_3P^+(CH_2)_3Se^-$ in solution, we have attempted to trap the latter by alkylation, to give 3-(methylseleno)propyl-triphenylphosphonium iodide (**6**). This was achieved by sodium borohydride reduction of the salt (**5a**), and alkylation of the resulting phosphonioalkylselenolate zwitterion with iodomethane to form **6** (Scheme 2).

Other workers have shown that alkyl selenocyanates also undergo reduction on treatment with sodium borohydride to form the related alkyl selenols [21].

NMR spectroscopy and electrospray mass spectrometry supported the formulation of this compound. When studied by MALDI TOFMS in positive ion mode (accurate mass analysis), an ion corresponding to the methylselenopropylphosphonium cation was observed. We therefore attempted to synthesize monolayer-protected cationic gold nanoparticles using the bis(triphenylphosphoniopropyl)diselenide (**5a**) and 6-(selenocyano)hexyl-triphenylphosphonium selenocyanates (**3b**) as the protecting ligands, assuming that Se–Se cleavage occurs in the interaction with gold. The synthesis of the gold nanoparticles was carried out following the same method as we reported in our previous work [4], *via* reduction of potassium tetrachloroaurate in a biphasic medium (dichloromethane:water) with an excess of sodium borohydride [22,23]. A solution of the ligand was prepared in dichloromethane (DCM) and solid potassium tetrachloroaurate (Se/Au molar ratio, 1:1) was then added to the solution. This was vigorously stirred until the gold salt was totally dissolved. The reduction was carried out by adding dropwise a freshly prepared aqueous solution of sodium borohydride with vigorous stirring, under a nitrogen atmosphere. After 1 h, the stirring was stopped and dark blue particles of aggregated colloidal gold were observed at the bottom of the flask. No evidence of stable gold nanoparticle formation was observed in both cases (**5a** and **3b**). Following this result, the molar quantity of the ligand was increased and a Se/Au molar ratio of 2:1 was used in order to assure the complete stabilisation of all the gold nanoparticles in solution. However, after the reduction and 2 h of vigorous stirring, particles of aggregated colloidal gold were observed again at the bottom of the reaction vessel. Even with a further increase in the quantity of ligand, the formation of functionalised gold nanoparticles was not achieved. If stirring is continued overnight then the blue aggregates slowly re-dissolve producing a pale yellow coloured solution. The observation that neither **5a** or **3b** facilitates the formation



Scheme 2. Synthesis of 3-(methylseleno)propyl-triphenylphosphonium iodide (**6**).

of stabilised gold nanoparticles is obviously disappointing. We postulate that the selenocyanate ions present in both **5a** and **3b** may be a complicating factor and it is well known that the cyanide ion, and other pseudohalides, have very high affinities for gold and are widely used for the dissolution, recovery and recycling of gold metal [24]. Furthermore, it has been reported that treatment of gold(I) captopril (captopril = 1-[(2*S*)-3-mercapto-3-methylpropionyl]-L-proline) with KSeCN or selenourea in aqueous solution produces unstable species that readily undergo disproportionation and decomposition [25]. Selenocyanate complexes of gold(I) are comparatively rare and complexes of the type $[(R_3P)Au(SeCN)]$, formed through the reaction of $[(R_3P)AuCl]$ with KSeCN in a biphasic water/dichloromethane system were found to be less stable thermally, and to air and moisture, than their thiocyanate analogues [26].

3. Experimental

3.1. General

^1H and ^{31}P NMR spectra were obtained in $CDCl_3$ and in $CDCl_3$/DMSO mixture on a Bruker DMX 250 (250 MHz) spectrometer. Electrospray mass spectra were recorded using an Applied Biosystems "QStar-Pulsar-i" hybrid quadrupole time of flight LCMS–MS instrument. UV–visible spectra of aqueous colloidal solutions were obtained on an ATI UNICAM UV2 spectrometer. Analytical thin layer chromatography (TLC) was performed on Merck silica gel $60F_{254}$ plates using 90:10 dichloromethane:methanol as eluent system.

3.2. Syntheses of bis(3-triphenylphosphoniopropyl) diselenide–di(selenocyanate) (*5a*) and 6-(selenocyano) hexyl-triphenylphosphonium selenocyanate (*3b*)

The synthesis of **5a** and **3b** was performed by the reaction of the corresponding (bromoalkyl)triphenylphosphonium bromide (1 mol) with potassium selenocyanate (4 mol) in aqueous ethanol under nitrogen and heated under reflux for 6 h. Progress of the reaction was monitored by TLC, using 10% methanol:90% dichloromethane as a mobile phase. The resulting compound was obtained by dichloromethane extractions of the reaction mixture and initially purified by trituration with dry diethyl ether.

Crystallisation from DCM–ether gave: *bis(3-triphenylphosphoniopropyl)diselenide–di(selenocyanate)–KOH* (**5a**) as yellow crystals, mp 106–109 °C, Anal. Calc. for $C_{44}H_{42}N_2P_2Se_4 \cdot KOH$: C, 51.17; H, 4.20; N, 2.71. Found: C, 51.47; H, 4.56; N, 2.55%. ESMS 383 [1/2M+H$^+$], 410 [1/2M+Na$^+$], 460 [1/2M+Se], 804 [M+K$^+$]. $\delta^{31}P(CDCl_3) = 23.4$ and 23.5 ppm, $\delta^1H(CDCl_3) = 2.2$ (2H, m), 3.2 (2H, t), 3.7 (2H, m), 7.7–7.8 (15H, m) ppm. X-ray analysis confirmed the structure, and also confirmed the presence of 1 mol of KOH per mole of diselenide salt.

6-(Selenocyano)hexyl-triphenylphosphonium selenocyanate (**3b**) yellow oil, accurate MALDI TOFMS analysis: found 452.1045 [M+H$^+$]; Cation $C_{25}H_{27}NPSe$ requires 452.1040 [M+H$^+$]. $\delta^{31}P(CDCl_3) = 23.6$ ppm, $\delta^1H(CDCl_3) = 1.5$–1.8 (6H, m), 1.9 (2H, m), 3.1 (2H, t), 3.5 (2H, m), 7.6–7.8 (15H, m) ppm.

3.3. Synthesis of 3-(methylseleno)propyl-triphenylphosphonium iodide (*6*)

The alkylation of **5a** was carried out using the following method: bis(3-triphenylphosphoniopropyl)diselenide–di(selenocyanate) (0.5 mmol) was dissolved in 3 mL of methanol. A freshly prepared aqueous solution of sodium borohydride (2 mL, 5 mmol) was then added drop by drop to the reaction flask, in order to allow formation of the zwitterion $Ph_3P^+(CH_2)_3Se^-$. The mixture was stirred for 1 h at room temperature. The formation of 3-(methylseleno)propyl-triphenylphosphonium iodide was achieved by the reaction of **5a** and methyl iodide (0.3 mL, 5 mmol) under nitrogen and the mixture was stirred overnight at room temperature (Scheme 2). Progress of the reaction was monitored by TLC, using 10% methanol:90% dichloromethane as a mobile phase. The resulting mixture was extracted with dichloromethane, the nonaqueous phase was collected and after removing the solvent, the resulting compound was initially purified by trituration with dry diethyl ether.

3-(Methylseleno)propyl-triphenylphosphonium iodide (**6**), pale cream solid. 62% yield, mp 140 °C. Accurate MALDI TOFMS analysis: found 399.0761 [M$^+$]; Cation $C_{22}H_{24}PSe$

requires 399.0780[M^+]. δ^{31}P NMR ($CDCl_3$) = 24.06 ppm, δ ^{1}H NMR ($CDCl_3$) = 1.8 (3H, s), 2.8 (2H, t), 3.2 (2H, m), 3.9 (2H, m), 7.6–7.8 (15H, m) ppm.

4. X-ray crystallography study of bis(3-triphenylphosphoniopropyl)diselenide–di(selenocyanate) (5a)

Single crystal X-ray data for **5a** were collected in a Bruker Nonius KappaCCD mounted at the window of a molybdenum rotating anode; standard procedures were followed for both data reduction and structure solution. During refinement the potassium site was treated as split over four locations in close proximity, and the hydrogen of the OH was located from the difference map and then refined using restraints.

4.0.1. Crystal data

$C_{44}H_{43}KN_2OP_2Se_4$, $M_r = 1032.68$, $T = 120(2)$ K, monoclinic, space group $P2_1/n$, $a = 12.4500(2)$, $b = 18.3601(2)$, $c = 19.4363(3)$ Å, $\beta = 103.069(1)°$, $V = 4327.74(11)$ Å^3, ρ_{calc} 1.585 g cm^{-3}, $\mu = 3.597$ mm^{-1}, $Z = 4$, reflections collected: 50997, independent reflections: 7618 ($R_{int} = 0.0890$), final R indices [$I > 2\sigma I$]: $R_1 = 0.0424$, $wR_2 = 0.0925$, R indices (all data): $R_1 = 0.0748$. $wR_2 = 0.1029$.

4.1. Attempts to synthesize gold nanoparticles using bis(triphenylphosphoniopropyl)diselenide (*5a*) and 6-(selenocyano)hexyl-triphenylphosphonium selenocyanate (*3b*) as protecting ligands

A solution of ligand was prepared in DCM (0.12 mmol, 7 mmol L^{-1}; and 0.25 mmol, 14 mmol L^{-1}) and potassium tetrachloroaurate (0.12 mmol, 7 mmol L^{-1}) was then added to the solution. This was vigorously stirred until the gold salt was totally dissolved. The reduction was carried out by adding dropwise a freshly prepared aqueous solution of sodium borohydride (3 mL, 400 mmol L^{-1}) with vigorous stirring, and 15 mL of deionised water was then added to the mixture.

Supplementary material

CCDC 635855 contains the supplementary crystallographic data for **5a**. These data can be obtained free of charge via http://www.ccdc.cam.ac.uk/conts/retrieving.html, or from the Cambridge Crystallographic Data Centre, 12 Union Road, Cambridge CB2 1EZ, UK; fax: (+44) 1223-336-033; or e-mail: deposit@ccdc.cam.ac.uk.

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