

A Synthesis of 6-(2,5-Dimethoxy-4-(2-aminopropyl)phenyl)-hexylthiol. A Ligand for Conjugation with Fluorescent Cadmium Selenide/Zinc Sulfide Core/Shell Nanocrystals and Biological Imaging.

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Received 13 August 2002; in revised form: 27 November 2002 / Accepted: 27 November 2002 / Published: 30 November 2002

Abstract: The synthesis of 6-(2,5-dimethoxy-4-(2-aminopropyl)phenyl)hexylthiol, an agonist with a very high affinity for the 5HT_{2A} serotonin receptor subtype is reported. This agonist was designed to be attached to highly fluorescent cadmium selenide/zinc sulfide core/shells via a thiol at the end of a linker arm. This conjugate has applications in biological assays and biological imaging.

Keywords: Cadmium selenide / zinc sulfide / core/shells / 5HT_{2A} / fluorescent imaging.

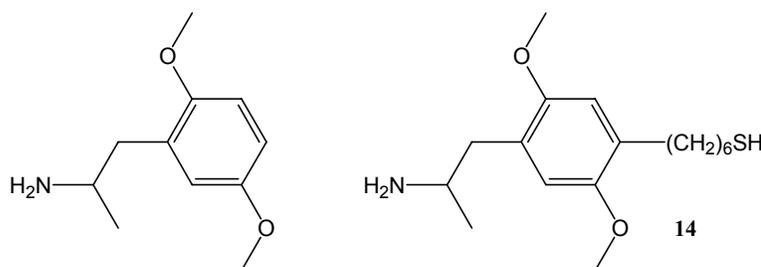
Introduction

When a narrow band gap semiconductor nanocrystal (or quantum dot) is coated with a shell of a wider band gap semiconductor, a new material, a core/shell nanocrystal, is produced. An example are CdSe/ZnS core/shell nanocrystals [1]. These core/shells have novel physical characteristics, one of the most important being size-tunable, narrow fluorescence emission bands. The fluorescence emission is a result of radiative recombination of the quantum confined electron-hole pair within the CdSe core. The core ranges in diameter from 18Å to 70Å and controls the emission wavelength. Small cores emit in the blue, while large cores emit in the red. The shell is several monolayers thick and serves both to

passivate dangling bonds on the core surface (these act as traps for the electron and hole and reduce the fluorescence quantum yield [2]) and to confine the photoexcited electron-hole pair to the core. Fluorescent CdSe/ZnS core/shells offer many distinct advantages over conventional dye molecules. They exhibit greater photostability and are not easily photobleached. Their quantum yields are comparable to or greater than organic dyes, and their absorption spectra are continuous above the first excitation feature, enabling all sizes of nanocrystals, and hence all colors, to be excited with a single excitation source. We are involved in utilizing the optical properties of core/shell nanocrystals for both static and dynamic fluorescent imaging of biological systems. Several groups have demonstrated that proteins and antibodies may be attached to core/shells [3-5], and these nanoconjugates have been shown to be capable of labeling specific cellular components. The narrow emission spectra of core/shell nanocrystals enables multiplexing experiments, where different biomolecules or drugs conjugated to different sizes of quantum dots can target multiple cellular components, and their distribution and dynamics can be visually observed. There is considerable effort being invested in optimizing fluorescent quantum dots for biological labeling systems [6]. It is hoped that such systems may find applications in genomics, proteomics, and high-throughput screening [7]

Our group is interested both in compounds that have biological activity in the central nervous system (CNS) and in synthesizing drug-core/shell conjugates that can be used to image receptor proteins, ion channels, and transporter proteins in neurons. This paper describes the synthesis and characterization of a ligand that has high affinity for the 5HT_{2A} and 5HT_{2C} serotonin receptors.[8] These receptors are G protein coupled receptors [9], and 5-HT_{2A} receptors have been linked to a wide range of behaviors and physiological functions including sleep, memory, hallucinations, anxiety, and aggression, as well as psychotic and affective diseases such as schizophrenia and unipolar depression. Our synthetic strategy is based upon attaching a flexible linker arm to the drug and attaching the other end to Zn atoms of the nanocrystal via a thiol group. We have already demonstrated that similar nanoconjugates are biologically active and can be used to image serotonin transporter proteins transfected in HEK cells [10]. 1-(2-Aminopropyl)-2,5-dimethoxybenzene (Figure 1) has been shown to have a high affinity for the 5HT₂ receptors. Recent publications indicated that there is a region in the molecule where increasing steric bulk is tolerated, and substituents with a large steric bulk have little or no detrimental effect on its biological activity [8,11]. This position is *para* to the isopropylamine; consequently we decided to attach an alkyl chain to this position giving the ligand 6-(2,5-dimethoxy-4-(2-aminopropyl)phenyl)hexylthiol (**14**).

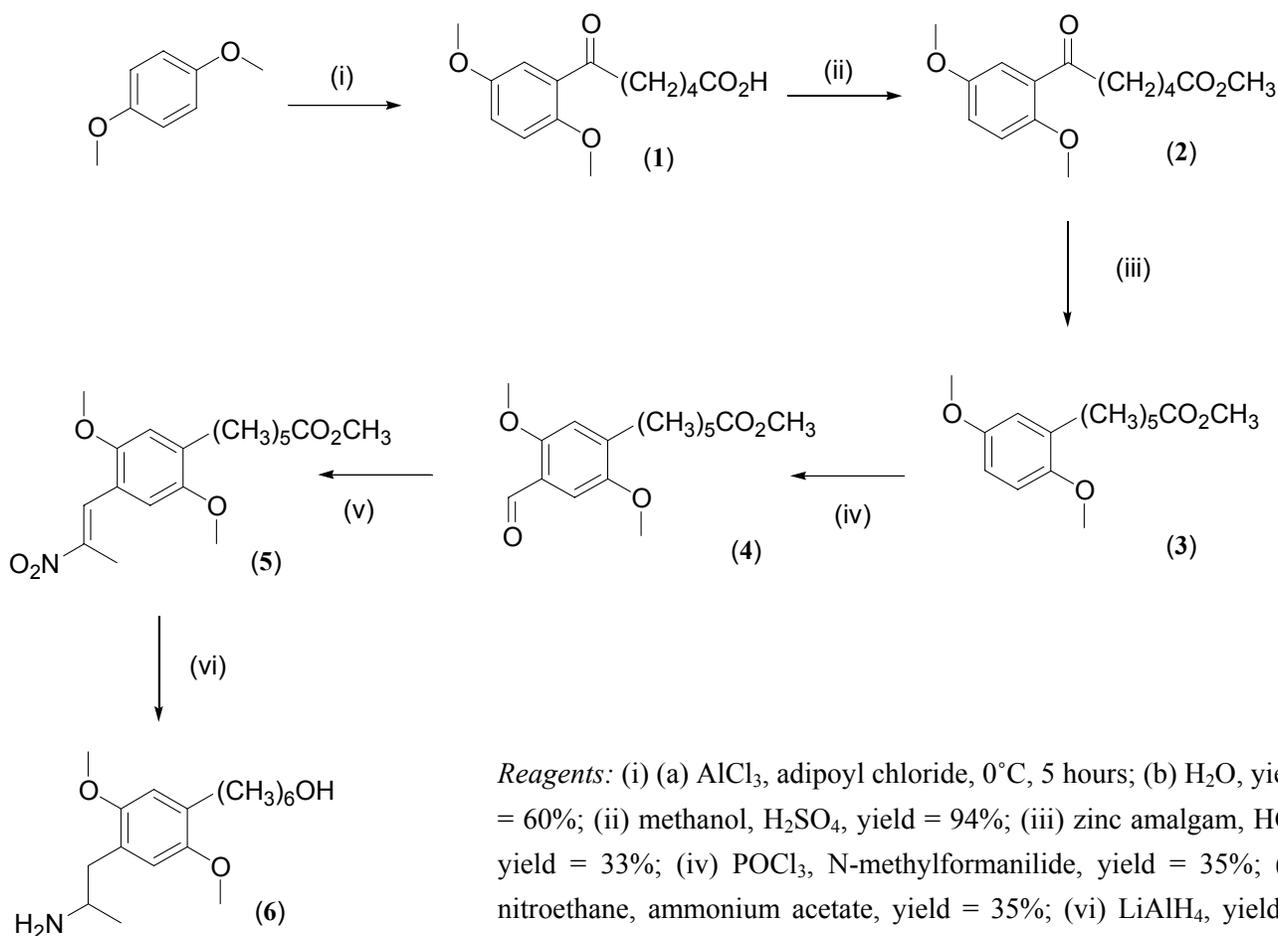
Figure 1. 1-(2-aminopropyl)-2,5-dimethoxybenzene, a known high affinity agonist for 5HT_{2A} receptors, and the structure of our proposed ligand **14**.



Results and Discussion

Several synthetic methodologies were considered in our design of the synthesis of 6-(2,5-dimethoxy-4-(2-aminopropyl)phenyl)hexylthiol (**14**). Our main criterion when selecting a route was to develop a reproducible method that used inexpensive, commercially available reagents. First the intermediate 1-(2,5-dimethoxy-4-(6-hydroxyhexyl))-2-aminopropane (**6**) was synthesized (Scheme 1), and this was then converted to 6-(2,5-dimethoxy-4-(2-aminopropyl)phenyl)hexylthiol via two different methods. Scheme 2 shows these methodologies.

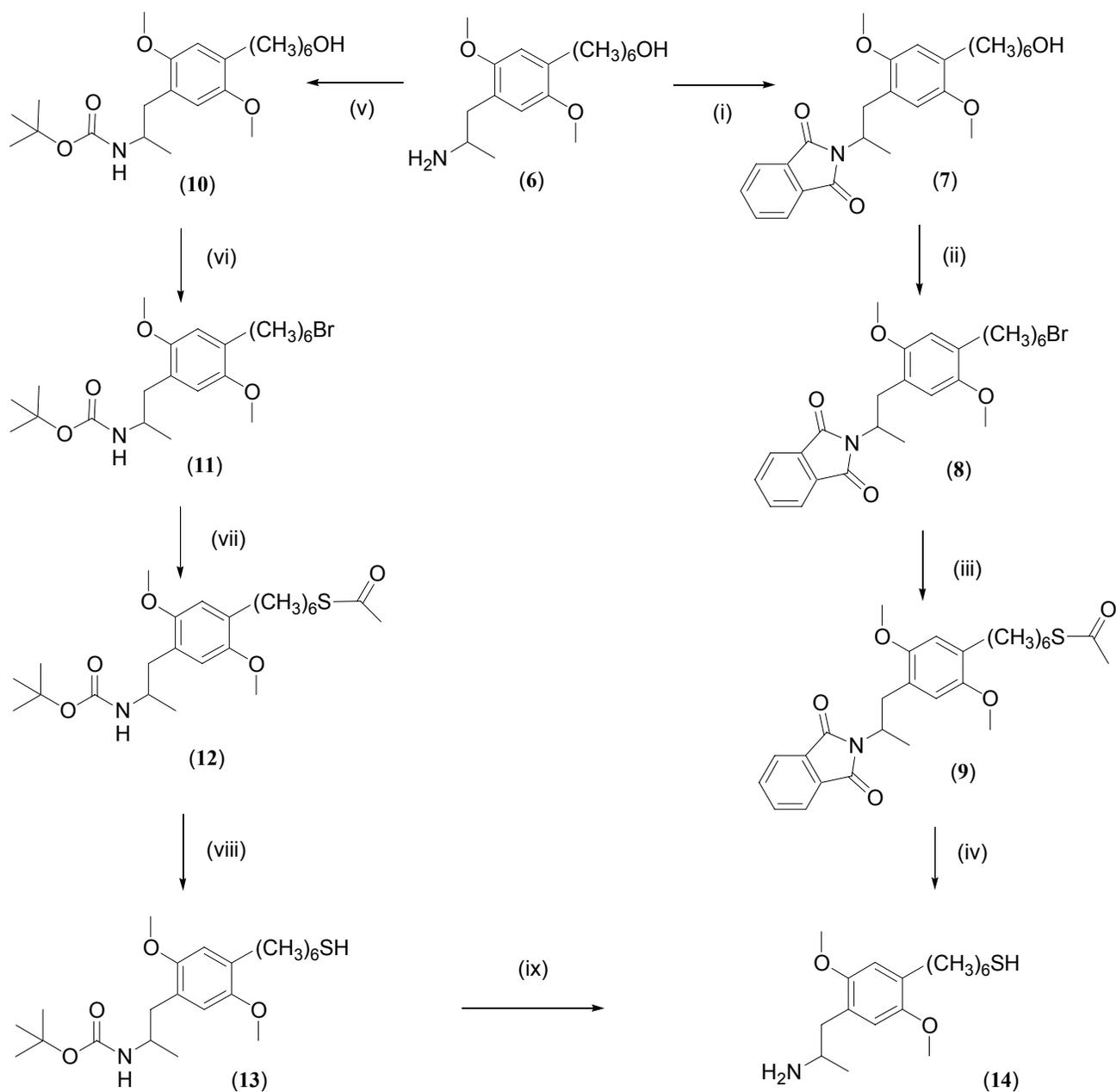
Scheme 1.



6-(2,5-Dimethoxyphenyl)-6-oxohexanoic acid (**1**) was synthesized by a Friedel-Crafts acylation of 1,4-dimethoxybenzene with adipoyl chloride. Hydrolysis of the resultant acid chloride gave **1** in 60% yield. Methyl 6-(2,5-dimethoxyphenyl)-6-oxohexanoate (**2**) was obtained by esterification of **1** in 94% yield and this was reduced using amalgamated zinc to give methyl 6-(2,5-dimethoxyphenyl)hexanoate (**3**) in 33% yield. A Vilsmeier-Haack formylation was performed on compound **3** giving methyl 6-(2,5-dimethoxy-4-formylphenyl)hexanoate (**4**) in 35% yield. The nitrostyrene **5** was obtained by refluxing

the aldehyde **4** with nitroethane in acetic acid. Methyl 6-(2,5-dimethoxy-4-(2-nitroprop-2-ene)phenyl)-hexanoate (**5**) was obtained as a yellow solid (35% yield) and reduced in the presence of 5 molar equivalents of lithium aluminum hydride to give 1-(2,5-dimethoxyphenyl-4-(6-hydroxyhexyl))-2-aminopropane (**6**) in 66% yield.

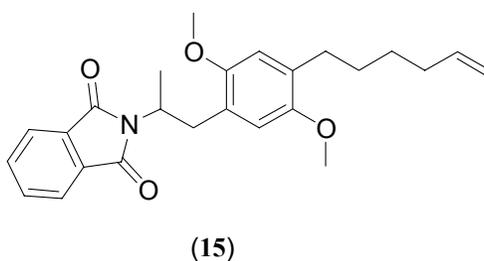
Scheme 2



Reagents: (i) N-carbonylphthalimide, yield = 95%; (ii) PPh₃, NBS, yield = 27%; (iii) potassium thioacetate, yield = 82%; (iv) hydrazine hydrate, yield = 68%; (v) *t*-BOC anhydride, yield = 47%; (vi) PPh₃, NBS, yield = 30%; (vii) potassium thioacetate, yield = 73%; (viii) ammonia, yield = 88%; (ix) trifluoroacetic acid, yield = 88%

The amine **6** was protected using either a phthalimido or a *t*-BOC protecting group, giving 6-(2,5-dimethoxy-4-(2-[N,N-phthalimido]propyl)phenyl)hexanol (**7**) and 6-(2,5-dimethoxy-4-(2-[N-(*tert*-butoxycarbonyl)]aminopropyl)phenyl)hexanol (**10**) in yields of 95% and 47%, respectively. The alcohols were then converted to protected thiols and the protecting groups were removed giving 6-(2,5-dimethoxy-4-(2-aminopropyl)phenyl)hexylthiol (**14**). Initially the alcohol was converted to a group that could be displaced with a sulfur-containing nucleophile such as a chloride, tosylate or bromide. Conversion to a chloride using thionyl chloride gave low yields of the chloride. Unwanted side products such as **15** (Figure 2) predominated; this was also observed when the alcohol was converted to a tosylate.

Figure 2. A potential side product formed during the synthesis of intermediates such as the chloro analogue of **8**.



When the alcohol was converted to a bromide using triphenyl phosphine and N-bromosuccinimide low yields of product were obtained. However it was possible to reclaim and reuse unreacted starting material giving 6-(2,5-dimethoxy)-4-(2-[N,N-phthalimido]propyl)phenyl)hexyl bromide (**8**) in a yield of 27% and a 30% yield of 6-(2,5-dimethoxy-4-(2-[N-(*tert*-butoxycarbonyl)]aminopropyl)phenyl)hexyl bromide (**11**). Nucleophilic substitution of bromine in compounds **8** and **11** with potassium thioacetate gave an 82% yield 6-(2,5-dimethoxy-4-(2-[N,N-phthalimido]propyl)phenyl)hexyl thioacetate (**9**) whilst 6-(2,5-dimethoxy-4-(2-[N-(*tert*-butoxycarbonyl)]aminopropyl)phenyl)hexyl thioacetate (**12**) was obtained in 73% yield.

Thioacetates are hydrolyzed under basic conditions giving thiols [12]. We were able to hydrolyze thioesters with hydrazine hydrate enabling the removal of two protecting groups in one pot from compound **9** (Method A, Experimental). 6-(2,5-Dimethoxy-4-(2-aminopropyl)phenyl)hexyl thiol (**14**) was obtained as a colorless oil in a yield of 68% by this method. Alternatively, the thioacetate was removed from compound **12** by hydrolysis in methanolic ammonia giving 6-(2,5-dimethoxy-4-(2-[N-(*tert*-butoxycarbonyl)]aminopropyl)phenyl)hexyl thiol (**13**) in 88% yield. *t*-BOC was subsequently removed in the final step using trifluoroacetic acid giving 6-(2,5-dimethoxy-4-(2-aminopropyl)phenyl)hexyl thiol (**14**) in 70% yield (Method B, Experimental). Compound **14** was purified by washing with hexanes and crystallization from ether as the oxalate salt. It was characterized by ¹H-NMR, ¹³C-NMR, low resolution mass spectroscopy and elemental analysis.

Conclusions

A synthetic route for the synthesis of 6-(2,5-dimethoxy-4-(2-aminopropyl)phenyl)hexylthiol (**14**) has been developed. This compound has been shown to be a full agonist with an EC₅₀ for the 5HT_{2A} receptor of 88 nM [13]. Conjugates of this compound with highly fluorescent cadmium selenide/zinc sulfide core/shells have been demonstrated to be biologically active and have been used in biological fluorescence studies [13].

Acknowledgements

We would like to thank Professor Ned Porter for a critical reading of this manuscript and Vanderbilt University for supporting this research in part by providing an intramural discovery grant. We would also like to acknowledge the National Institute of Health for provision of grant # 5R03191-161874-02.

Experimental

General

Adipic acid, N-bromosuccinimide, potassium thioacetate, N,N-phthalimido-2-(5-hydroxy-1H-indole-3-yl)ethylamine, potassium thioacetate and triphenyl phosphine were purchased from Aldrich. 1,4-Dimethoxybenzene, aluminum chloride, and lithium aluminum hydride were purchased from Lancaster Synthesis. Thionyl chloride was purchased from ACROS. Reagents were used as they were received. Thin-layer chromatography was carried out on pre-coated plates, and the products were visualized using UV light. Column chromatography on silica refers to silica gel obtained from Scientific Adsorbents, Inc., catalogue number 02826-25. All NMR data was obtained using a Bruker 300 MHz machine with CDCl₃ as a solvent unless otherwise stated. Chemical shifts were measured in ppm relative to TMS and coupling constants are measured in Hz. GC-MS was performed on a Hewlett Packard 5890 series II gas chromatogram coupled to a Hewlett Packard 5971 mass spectroscope using electron impact as the ionization method. Low resolution mass spectra (MS) were obtained using a Finnegan Thermoquest TSQ 7000 triple quadrupole LC-MS equipped with an API-1 electrospray ionization source (ES). Samples for elemental analysis were routinely dried at ca. 10mmHg. Elemental analysis was performed by Atlantic Microlabs, Georgia, and the analysis is corrected for hydrates when necessary.

6-(2,5-Dimethoxyphenyl)-6-oxohexanoic acid (**1**)

Adipoyl chloride (50 mL) and aluminum chloride (10g, 7.4 mmols) were dissolved in nitrobenzene (50 mL) and cooled to 0°C. 1,4-Dimethoxybenzene (10g, 7.2 mmols) dissolved in nitrobenzene (50 mL) was added dropwise over a 3 hour period, during which the temperature was

maintained below 5°C. The resulting mixture was stirred for a further 2 hours at 0°C, then crushed ice was added. The reaction mixture was allowed to warm to room temperature over an 18 hour period and the solution was filtered. The organic solution was separated and extracted into 3M sodium hydroxide solution (3 x 100 mL) and the aqueous solution was acidified to pH 1 using 4M hydrochloric acid. After extracting this solution with diethyl ether (3 x 200 mL) the ethereal extracts were combined and dried over magnesium sulfate. The solution was filtered and evaporated giving the crude product as a brown solid. It was purified by recrystallization from a mixture of ethyl acetate and hexane. This gave the product 11.4g (60%) as a colorless solid $R_f = 0.38$ (silica, 9:1 dichloromethane-methanol); m.p. = 75-77°C (lit. [14] = 78-80°C); $^1\text{H-NMR}$ (DMSO- d_6) δ 1.50 - 1.55 (m, 4H), 2.22 (t, $J = 3.7$ Hz, 2H), 2.91 (t, $J = 7.0$ Hz, 2H), 3.73 (s, 3H), 3.82 (s, 3H), 7.05 (t, $J = 1.8$ Hz, 1H), 7.09 (d, $J = 1.8$ Hz, 2H), 11.99 (s, 1H); $^{13}\text{C-NMR}$ (DMSO- d_6) δ 23.75, 24.50, 33.72, 33.88, 55.86, 56.58, 113.85, 114.25, 119.23, 128.99, 152.50, 153.27, 174.66, 174.70

Methyl 6-(2,5-dimethoxyphenyl)-6-oxohexanoate (2)

6-(2,5-Dimethoxyphenyl)-6-oxohexanoic acid (**1**, 4.2g, 160 mmols) was added to a mixture of methanol (100 mL) and concentrated sulfuric acid (2 drops). The solution was heated at reflux over a period of 18 hours with stirring. After cooling to room temperature the solution was evaporated and the crude product was dissolved in diethyl ether (100 mL). This was washed with saturated sodium bicarbonate (50 mL) and water (50 mL). It was dried over magnesium sulfate, filtered and evaporated. The product was purified using column chromatography on silica gel (dichloromethane elution). This gave 4.2g (94%) of the product as a pale yellow oil; $R_f = 0.24$ (silica, dichloromethane); $^1\text{H-NMR}$: δ 1.39 – 1.42 (m, 4H), 2.17 (t, $J = 6.9$ Hz, 2H), 2.69 (t, $J = 6.8$ Hz, 2H), 3.51 (s, 3H), 3.70 (s, 3H), 3.85 (s, 3H), 6.61 (d, $J = 9.0$ Hz, 1H), 6.70 (dd, $J = 9.0$ Hz and $J = 1.0$ Hz, 1H), 6.93 (d, $J = 3.0$ Hz, 1H); $^{13}\text{C-NMR}$: δ 23.40, 24.21, 33.13, 33.44, 50.94, 55.21, 55.55, 112.69, 113.56, 119.11, 128.14, 152.53, 153.04, 173.13, 173.37; GC-MSEI [M^+] 281, [M^+] 280; Calculated: C=64.22%, H=7.19%; found C=64.06%, H=7.03%

Methyl 6-(2,5-dimethoxyphenyl)hexanoate (3)

Powdered zinc (22.5g) was added to a solution of mercuric chloride (940 mg) in concentrated hydrochloric acid (0.93 mL) and water (23.1 mL). This suspension was shaken for 5 minutes and the liquid was decanted. The amalgamated zinc was placed in a 500ml 3 necked flask and concentrated hydrochloric acid (12 mL) was added. The flask was heated to cause a gentle reflux and a solution of methyl-6-(2,5-dimethoxyphenyl)-6-oxohexanoate (**2**, 4.2g, 15 mmols) in methanol (7 mL) and concentrated hydrochloric acid (23 mL) was added dropwise [15]. The mixture was heated at reflux for 3 hours following the addition of compound **2**, then filtered. The aqueous solution was extracted with diethyl ether (4 x 100 mL) and the combined ethereal extracts were washed with saturated sodium bicarbonate (50 mL) and water (50 mL). After drying over magnesium sulfate the solution

was filtered and evaporated. The product was purified by column chromatography on silica gel (98:2 dichloromethane-methanol). This gave 1.35g (33%) of the product as a pale yellow oil; $R_f = 0.67$ (silica, 98:2 dichloromethane-methanol); $^1\text{H-NMR}$: δ 1.26- 1.41 (m, 2H), 1.53 - 1.76 (m, 4H), 2.30 (t, $J = 7.6$ Hz, 2H), 2.57 (t, $J = 7.8$ Hz, 2H), 3.54 (s, 3H), 3.74 (s, 3H), 3.75 (s, 3H), 6.64 - 6.78 (m, 3H); $^{13}\text{C-NMR}$: δ 24.65, 28.79, 29.19, 29.85, 33.75, 51.09, 55.29, 55.72, 110.41, 110.96, 115.96, 131.93, 151.53, 153.24, 173.90; GC-MSEI [M^+] 267, [M^+] 266; Calculated: C = 67.67%, H = 8.33%; found C = 67.91%, H = 8.15%

Methyl 6-(2,5-dimethoxy-4-formylphenyl)hexanoate (4).

A mixture of phosphorus oxychloride (1mL) and N-methylformanilide (1.81 g) were allowed to incubate at room temperature for 30 minutes. Methyl-6-(2,5-dimethoxyphenyl)hexanoate (**3**, 1 g, 4 mmols) was added and the mixture was heated for 2 hours. After cooling to room temperature water (50 mL) was added and the mixture was left standing at room temperature for 18 hours. Then the solution was extracted with dichloromethane (2 x 100mL) dried over magnesium sulfate, filtered and evaporated. The resulting oil was leached with boiling hexanes (4 x 100mL) and the combined solutions were evaporated under reduced pressure [16]. Purification of the product was accomplished by column chromatography on silica gel (98:2 dichloromethane-methanol). This gave 0.4g (35%) of the product as a colorless solid; $R_f = 0.25$ (silica, 95:5 dichloromethane-methanol); m.p. = 74-76°C; $^1\text{H-NMR}$: δ 1.38 (m, 2H), 1.60 (m, 4h), 2.32 (t, $J = 7.5$ Hz, 2H), 2.64 (t, $J = 7.9$ Hz, 2H), 3.67 (s, 3H), 3.82 (s, 3H), 3.89 (s, 3H), 6.79 (s, 1H), 7.27, (s, 1H), 10.40 (s, 1H); $^{13}\text{C-NMR}$: δ 22.10, 26.35, 26.54, 28.25, 48.86, 53.16, 53.58, 105.49, 111.23, 120.30, 138.13, 149.08, 154.10, 171.56, 186.61; GC-MSEI [M^+] 295, [M^+] 294; Calculated: C = 65.53%, H = 7.50%; found C = 65.30%, H = 7.55%

Methyl 6-(2,5-dimethoxy-4-(2-nitroprop-2-ene)phenyl)hexanoate (5).

Methyl 6-(2,5-dimethoxy-4-formylphenyl)hexanoate (**4**, 1g, 3.4 mmols) was added to glacial acetic acid (100mL). This was followed by ammonium acetate (272 mg) and nitroethane (1mL). The mixture was heated at reflux for 4 hours and then it was evaporated. The product was purified by column chromatography on silica gel (25:75 ethyl acetate-hexanes). This gave 420 mg (35%) of the product as a yellow solid; $R_f = 0.65$ (silica, 1:1 ethyl acetate-hexanes); m.p. = 57-58°C; $^1\text{H-NMR}$: δ 1.37 (q, $J = 6.5$ Hz, 2H), 1.57 - 1.73 (m, 4H), 2.32 (t, $J = 7.5$ Hz, 2H), 2.41 (s, 3H), 2.63 (t, $J = 7.8$ Hz, 2H), 3.65 (s, 3H), 3.80 (s, 3H), 3.90 (s, 3H), 6.77 (s, 1H), 6.79 (s, 1H), 8.18 (s, 1H); $^{13}\text{C-NMR}$: δ 24.35, 28.57, 28.87, 30.05, 33.49, 50.93, 55.55, 55.64, 111.48, 112.73, 118.44, 129.42, 135.06, 146.31, 150.78, 152.13, 173.64; Calculated: C = 61.52%, H = 7.17%, N = 3.99%; found C = 61.66%, H = 7.07%, N = 4.09%

1-(2,5-Dimethoxy-4-(6-hydroxyhexyl))-2-aminopropane (6).

Methyl 6-(2,5-dimethoxy-4-(2-nitroprop-2-ene)phenyl)hexanoate (**5**, 420 mg, 1.2 mmols) was dissolved in dry diethyl ether (100 mL) and a solution of lithium aluminum hydride (1M, 14 mL) was added. The mixture was heated at reflux for 48 hours under nitrogen then stirred at room temperature for a further 2 days under nitrogen. The solution was cooled to 0°C in an ice-acetone bath and sulfuric acid (8%) was added until hydrogen evolution ceased. The aqueous solution was separated and washed with diethyl ether (2 x 50 mL). The aqueous solution was basified with sodium bicarbonate to pH 8 and the aluminum salts were removed by filtration. The inorganic salts were dried and washed with dichloromethane (2 x 100mL) and the aqueous solution was extracted with dichloromethane (2 x 100mL). The combined organic extracts were dried over magnesium sulfate, filtered, and evaporated to yield 250 mg (66%) of the product as a colorless solid. The product was converted to the oxalate salt by dissolving the base (20 mg, 0.68 mmols) in dry ether and adding oxalic acid (100 mg, 1.1 mmols). The resulting oxalate salt was recrystallized from isopropanol giving 10 mg (38%) of the product as a colorless solid, m.p. = 164-166°C; Calculated (oxalate salt + 1/4 H₂O): C = 58.52%, H = 8.14% , N = 3.59%; found C = 58.69%, H = 8.00%, N = 3.66%. The oxalate salt was converted to the base for the next step in the synthetic route; ¹H-NMR: δ 1.10 (d, *J* = 4.7 Hz, 3H), 1.37 - 1.38 (m, 4H), 1.51 - 1.59 (m, 4H), 2.46 - 2.70 (m, 7H), 3.20 (q, *J* = 5.4 Hz , 1H), 3.56 (t, *J* = 4.9 Hz, 2H), 3.74 (s, 3H), 3.75 (s, 3H), 6.63 (s, 1H), 6.66 (s, 1H); ¹³C-NMR: δ 20.90, 25.22, 28.84, 29.56, 32.23, 40.14, 46.62, 55.06, 55.24, 55.39, 61.11, 112.29, 113.42, 124.96, 129.32, 150.52, 150.91

6-(2,5-Dimethoxy-4-(2-[N,N-phthalimido]propyl)phenyl)hexanol (7).

1-(2,5-Dimethoxy-4-(6-hydroxyhexyl))-2-aminopropane (**6**, 250 mg, 0.8 mmols) was dissolved in tetrahydrofuran (10mL). A solution of sodium bicarbonate (100 mg) in water (10mL) was added followed by N-carbethoxyphthalimide (175 mg, 0.8 mmols) The mixture was stirred at room temperature for 18 hours and then extracted with dichloromethane (2 x 50mL). The combined organic extracts were washed with water (20mL) dried over magnesium sulfate, filtered and evaporated under reduced pressure. The product was purified by column chromatography on silica (1:1 ethyl acetate-hexanes). This gave 0.326g (95%) of the product as a colorless solid, m.p. = 82-83 °C; *R_f* = 0.4 (silica, 1:1 ethyl acetate-hexanes); ¹H-NMR: δ 1.28 - 1.56 (m, 11H), 1.73 (br s, 1H), 2.48 (t, *J* = 7.7 Hz, 2H), 3.14 (m, 2H), 3.52 (s, 3H), 3.60 (t, *J* = 6.4 Hz, 2H), 3.71 (s, 3H), 4.65 - 4.77 (m, 1H), 6.52 (s, 1H), 6.55 (s, 1H), 7.62 - 7.66 (m, 2H), 7.71 - 7.75 (m, 2H); ¹³C-NMR: δ 18.40, 25.43, 28.99, 29.84, 29.91, 32.57, 34.73, 47.30, 55.79, 55.90, 62.76, 112.61, 113.69, 122.72, 124.49, 130.06, 131.90, 133.56, 150.84, 151.43, 168.42; Calculated: C = 70.57%, H = 7.34% , N = 3.29%; found: C = 70.51%, H = 7.34%, N = 3.25%.

6-(2,5-Dimethoxy-4-(2-[N,N-phthalimido]propyl)phenyl)hexyl bromide (8)

6-(2,5-Dimethoxy-4-(2-[N,N-phthalimido]propyl)phenyl)hexanol (**7**, 200 mg, 0.4 mmols) was dissolved in dichloromethane (20 mL) and cooled to 0°C. Triphenylphosphine (130 mg, 0.51 mmols) was dissolved in dichloromethane (10 mL) and added drop wise to the solution of **7**. After stirring for 30 minutes a solution containing N-bromosuccinimide (90 mg, 0.5 mmols) in dichloromethane (10 mL) was added dropwise over 10 minutes. The solution was stirred for 10 minutes at 0°C after the addition of N-bromosuccinimide was complete. Then it was allowed to warm to 22°C and it was stirred for 2 hours at this temperature [17]. The solvent was removed under reduced pressure and the product was purified by column chromatography on silica (1:1 ethyl acetate-hexanes). This gave 60 mg (27%) of the title compound as a yellow oil; $R_f = 0.65$ (silica, 1:1 ethyl acetate-hexanes); $^1\text{H-NMR}$: δ 1.26 – 1.56 (m, 7H), 1.83 (t, $J = 7.4$ Hz, 2H), 2.48 (t, $J = 7.5$ Hz, 2H), 3.05–3.28 (m, 4H), 3.38 (t, $J = 6.8$ Hz, 2H), 3.53 (s, 3H), 3.73 (s, 3H), 4.66 – 4.78 (m, 1H), 6.53 (s, 1H), 6.55 (s, 1H), 7.63 – 7.75 (m, 4H); $^{13}\text{C-NMR}$: δ 18.45, 27.94, 28.42, 29.73, 29.93, 32.72, 33.98, 34.86, 47.29, 55.86, 55.91, 112.65, 113.69, 122.74, 124.61, 129.90, 131.98, 133.59, 150.87, 151.48, 168.41; Calculated: C = 61.48%, H = 6.19%, N = 2.87%; found C = 61.43%, H = 6.20%, N = 2.79%

6-(2,5-Dimethoxy-4-(2-[N,N-phthalimido]propyl)phenyl)hexyl thioacetate (9).

6-(2,5-Dimethoxy-4-(2-[N,N-phthalimido]propyl)phenyl)hexyl bromide (**8**, 280 mg, 0.57 mmols) was dissolved in dry dimethylformamide (10 mL) and 4 Å molecular sieves (10 pellets) were added. The solution was stirred for 1 hour at room temperature and the potassium thioacetate (130 mg, 1.14 mmols) was added. The solution was stirred at room temperature for 18 hours. Then it was filtered and diethyl ether (100 mL) was added to the solution. The organic solution was washed with water (2 x 20mL), 1M hydrochloric acid (1 x 20mL), water (2 x 20 mL), 0.1M sodium bicarbonate (1 x 20mL). Then the ethereal solution was dried over magnesium sulfate filtered and evaporated under reduced pressure. The product was purified by column chromatography on silica (33:77 ethyl acetate-hexanes). This gave 230 mg (82%) of the product as a pale yellow oil; $R_f = 0.6$ (silica, 1:1 ethyl acetate-hexanes); $^1\text{H-NMR}$: δ 1.25 – 1.56 (m, 7H), 2.30 (s, 3H), 2.47 (t, $J = 7.0$ Hz, 2H), 2.86 (t, $J = 7.4$ Hz, 2H), 3.05 – 3.11 (m, 4H), 3.20–3.28 (m, 2H), 3.52 (s, 3H), 3.72 (s, 3H), 4.68 – 4.73 (m, 1H), 6.53 (s, 1H), 6.56 (s, 1H), 7.62 – 7.74 (m, 4H); $^{13}\text{C-NMR}$: δ 18.34, 28.47, 28.68, 28.97, 29.29, 29.66, 29.84, 30.44, 34.67, 47.16, 55.71, 55.76, 112.52, 113.55, 122.61, 124.42, 129.87, 131.86, 133.47, 150.75, 151.35, 161.25, 195.72; Calculated: C = 67.06%, H = 6.88%, N = 2.90%, S = 6.63%; found C = 66.72%, H = 6.96%, N = 2.97%, S = 6.58%

6-(2,5-Dimethoxy-4-(2-[N-(tert-butoxycarbonyl)]aminopropyl)phenyl)hexanol (10).

1-(2,5-Dimethoxy-4-(6-hydroxyhexyl))-2-aminopropane (**6**, 250 mg, 0.085 mmols) was dissolved in methanolic hydrochloric acid (30 mL) and this was evaporated. Once all the methanol had been

removed the resulting solid was dissolved in water (10 mL) and potassium carbonate (250 mg) was added all at once followed by di-*t*-butyl dicarbonate (200 mg, 1.1 mmols). The mixture was stirred at room temperature overnight and then extracted with dichloromethane (3 x 50 mL). The combined organic extracts were dried over magnesium sulfate, filtered, and evaporated. The product was purified by column chromatography on silica (95:5 dichloromethane-methanol). This gave 18 mg (47%) of the product as a colorless solid; $R_f = 0.65$ (silica, 97: 3 dichloromethane-methanol); $^1\text{H-NMR}$: δ 1.12 (d, $J = 4.8$ Hz, 3H), 1.34 (s, 9H), 1.55-1.58 (m, 6H), 2.21-2.33 (m, 2H), 2.68 (t, $J = 6.0$ Hz, 2H), 2.73-2.83 (m, 2H), 3.61 (t, $J = 6.0$ Hz, 2H), 3.76 (s, 3H), 3.78 (s, 3H), 4.86 (s, 1H), 6.63 (s, 1H), 6.65 (s, 1H); $^{13}\text{C-NMR}$: δ 21.33, 26.03, 28.81, 29.76, 30.49, 30.58, 33.10, 37.03, 47.91, 56.43, 63.17, 79.07, 113.19, 114.38, 125.07, 130.38, 151.58, 151.75, 155.84; Calculated: C = 66.80%, H = 9.43%, N = 3.54%; found C = 66.62%, H = 9.39%, N = 3.56%

6-(2,5-Dimethoxy-4-(2-[N-(tert-butoxycarbonyl)]aminopropyl)phenyl)hexyl bromide (11).

6-(2,5-Dimethoxy-4-(2-[N-(tert-butoxycarbonyl)]aminopropyl)phenyl)hexanol (**10**, 180 mg, 0.45 mmols) was dissolved in dichloromethane (20 mL) and cooled to 0°C. Triphenylphosphine (130 mg, 0.49 mmols) in dichloromethane (10 mL) was added dropwise followed by N-bromosuccinimide (90 mg, 0.5 mmols) in dichloromethane (10 mL). The solution was stirred at 0°C for 5 minutes following the addition of N-bromosuccinimide and then the solution was allowed to warm to 22°C. It was stirred at 22°C for 2 hours after which the dichloromethane was removed under reduced pressure and the product was purified using column chromatography on silica (1:1 ethyl acetate-hexanes). This gave 0.07g (30%) of the product as a yellow oil; $R_f = 0.6$ (silica, 1:1 ethyl acetate-hexanes); $^1\text{H-NMR}$: δ 1.05 (d, $J = 6.0$ Hz, 3H), 1.31 (s, 9H), 1.32-1.53 (m, 4H), 1.74-1.81 (m, 2H), 2.50 (t, $J = 6.0$ Hz, 2H), 2.63 (t, $J = 6.0$ Hz, 2H), 3.33 (t, $J = 6.5$ Hz, 2H), 3.69 (s, 3H), 3.71 (s, 3H), 3.77-3.80 (m, 1H), 6.56 (s, 1H), 6.58 (s, 1H), 6.70 (br s, NH); $^{13}\text{C-NMR}$: δ 20.86, 27.95, 28.35, 28.58, 29.82, 30.07, 32.71, 33.94, 36.64, 47.49, 55.90, 55.99, 78.61, 112.74, 113.88, 124.73, 129.74, 151.15, 151.33, 155.34; Calculated: C = 57.64%, H = 7.92%, N = 3.06%; found C = 57.56%, H = 7.87%, N = 3.11%

6-(2,5-Dimethoxy-4-(2-[N-(tert-butoxycarbonyl)]aminopropyl)phenyl)hexyl thioacetate (12).

6-(2,5-Dimethoxy-4-(2-[N-(tert-butoxycarbonyl)]aminopropyl)phenyl)hexyl bromide (**11**, 70 mg, 0.15 mmols) was dissolved in dry dimethylformamide (2 mL) and 4 Å molecular sieves (6 pellets) were added. The mixture was stirred at 22°C for 1 hour, then potassium thioacetate (35 mg, 0.3 mmols) was added. The solution was stirred for 18 hours at 22°C, filtered and diethyl ether (50 mL) was added. This was washed with 0.1M hydrochloric acid (1 x 10 mL), water (2 x 10 mL), 0.1M sodium bicarbonate (1 x 10 mL) and water (1 x 10 mL). The organic solution was dried over magnesium sulfate, filtered and evaporated. Then the product was purified using column chromatography on silica (1:1 ethyl acetate-hexanes). This gave 51 mg (73%) of the title product as a colorless solid, m.p. = 91-92°C; $R_f = 0.6$ (silica, 1:1 ethyl acetate-hexanes); $^1\text{H NMR}$ (CDCl_3) δ 1.13

(d, $J = 5.5$ Hz, 3H), 1.38 (s, 11H), 1.47 – 1.59 (m, 6H), 2.32 (s, 3H), 2.55 (t, $J = 7.8$ Hz, 2H), 2.73 (t, $J = 6.2$ Hz, 2H), 2.88 (t, $J = 6.4$ Hz, 2H), 3.76 (s, 3H), 3.78 (s, 3H), 3.80 – 3.90 (m, 1H), 4.76 (s, 1H), 6.62 (s, 1H), 6.64 (s, 1H); ^{13}C NMR (CDCl_3) δ 21.48, 28.37, 28.64, 28.99, 29.11, 29.42, 29.90, 30.12, 30.58, 36.65, 47.51, 55.92, 56.03, 78.63, 112.76, 113.91, 124.69, 129.89, 151.17, 151.36, 155.37, 195.95; Calculated: C = 63.54%, H = 8.67%, N = 3.09%, S = 7.07%; found: C = 63.51%, H = 8.72%, N = 3.16%, S = 7.11%

6-(2,5-Dimethoxy-4-(2-[N-(tert-butoxycarbonyl)]aminopropyl)phenyl)hexyl thiol (13)

6-(2,5-Dimethoxy-4-(2-[N-(tert-butoxycarbonyl)]aminopropyl)phenyl)hexyl thioacetate (**12**, 51 mg, 0.11 mmols) was dissolved in methanol (5 mL) and methanolic ammonia (25 mL) was added. The mixture was stirred at 22°C for 3 hours and evaporated. This gave a yellow tar which was dissolved in dichloromethane (50mL), the organic solution was washed with water (1 x 20mL) and dried over magnesium sulfate. After filtering it was evaporated and purified using silica gel column chromatography (1:1 ethyl acetate-hexanes). This gave 40 mg (88%) of compound **13** as a colorless solid, m.p. = 99-100°C; $R_f = 0.57$ (silica, 1:1 ethyl acetate-hexanes); $^1\text{H-NMR}$: δ 1.05 (d, $J = 6.5$ Hz, 3H), 1.13 – 1.59 (m, 17H), 2.41 – 2.70 (m, 6H), 3.69 (s, 3H), 3.71 (s, 3H), 3.76- 3.77 (m, 1H), 4.69 (s, 1H), 6.55 (s, 1H), 6.58 (s, 1H); $^{13}\text{C-NMR}$: 20.44, 24.16, 27.94, 28.58, 28.69, 29.50, 30.15, 33.51, 38.64, 47.05, 55.49, 55.60, 78.23, 112.33, 113.48, 124.27, 129.45, 150.75, 150.93, 154.95; Calculated: C = 64.20%, H = 9.06%, N = 3.40%, S = 7.79%; found: C = 64.04%, H = 8.95%, N = 3.41%, S = 7.68%

6-(2,5-Dimethoxy-4-(2-aminopropyl)phenyl)hexyl thiol (14)

Method A: 6-(2,5-Dimethoxy-4-(2-[N,N-phthalimido]propyl)phenyl)hexyl thioacetate (**9**, 230 mg, 0.47 mmols) was dissolved in absolute ethanol (50 mL). Hydrazine monohydrate (15 mL) was added to this solution and the mixture was stirred at 22°C for 90 minutes. Dichloromethane (200 mL) was added and the solution was washed with water (2 x 100 mL). The organic solution was dried over magnesium sulfate, filtered, and evaporated. This gave 100 mg (68%) of the product as a pale yellow oil; $^1\text{H-NMR}$: δ 1.13 (d, $J = 5.9$ Hz, 3H), 1.23 (m, 1H), 1.40 (m, 6H), 1.57 (t, $J = 6.0$ Hz, 2H), 1.68 (t, $J = 6.0$ Hz, 2H), 2.40 (br s, 2H), 2.56 (t, $J = 6.0$ Hz, 2H), 2.68 (t, $J = 7.3$, 2H), 3.21 (m, 1H), 3.76 (s, 6.0 H), 6.65 (s, 2H); $^{13}\text{C-NMR}$: δ 23.26, 28.36, 29.97, 30.14, 39.11, 40.64, 47.30, 56.01, 56.13, 112.95, 114.06, 125.67, 129.82, 151.12, 151.49; LR-ESMS [M^+] 312.32; The base was converted to the corresponding oxalate salt by dissolving 50 mg of the amine in dry ether (50 mL) and adding an ethereal solution containing oxalic acid (100 mg) dissolved in ether (200 mL). The resulting product was removed by filtration, washed with dry ether (2 x 100 mL) and air dried to give 20 mg of the oxalate as a colorless solid, m.p. = 148-150°C; Calculated: C = 56.84%, H = 7.78%, N = 3.49%, S = 7.98% found: C = 57.03%, H = 7.63%, N = 3.36%, S = 7.78%.

Method B: 6-(2,5-Dimethoxy-4-(2-[N-(*tert*-butoxycarbonyl)]aminopropyl)phenyl)hexyl thiol (**13**, 41 mg, 0.97 mmols) was dissolved in dry toluene (10 mL) and trifluoroacetic acid (0.2 mL) was added. The mixture was stirred at 22°C for 1 hour and then the solvent was removed under reduced pressure. The resultant tar was dissolved in dichloromethane (20 mL) and this was washed with sodium bicarbonate (0.1M, 1 x 20 mL) and water (2 x 10 mL). After drying over magnesium sulfate the solution was filtered and evaporated. This gave 21 mg (70%) of the product as a pale yellow oil.

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Sample Availability: Samples of compounds **1**, **2** and **5** are available from MDPI.

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