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# Graphical Abstract

## A novel colorimetric and fluorescent sensor for cyanide anions detection based on triphenylamine and benzothiadiazole

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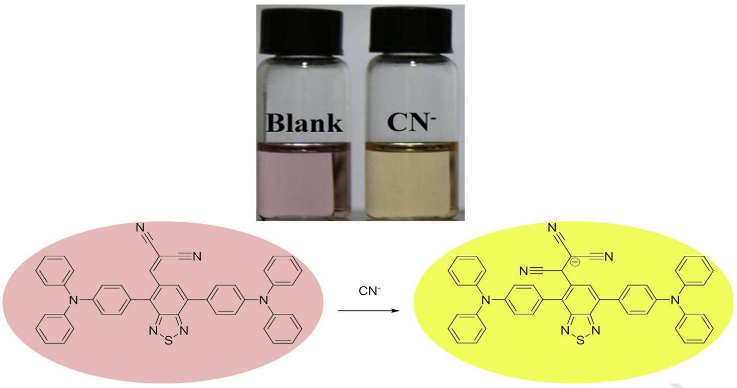

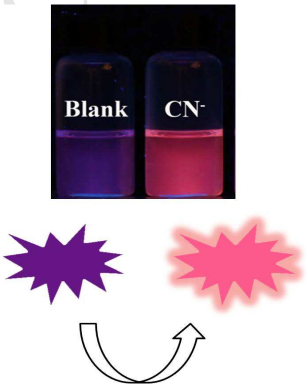

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ABSTRACT

A chemical sensor containing triphenylamine and benzothiadiazole for cyanide anions detection was designed and synthesized, whose structural feature is a donor-acceptor-donor (D-A-D) molecular configuration, which is substituted with a dicyanovinyl group served as a sensing unit. The nucleophilic addition of cyanide ion to the dicyanovinyl moiety was activated in the principle of Michael addition, which broken the electron-withdrawing interaction of dicyanovinyl moiety. The obstruction in the intramolecular charge transfer (ICT) induced remarkable changes in the absorption and emission spectra. The sensor exhibited splendid selectivity and anti-interference performance towards $CN^-$ over other anions ($F^-$, $Cl^-$, $Br^-$, $I^-$, $NO_2^-$ , $NO_3^-$, $HCO_3^-$, $H_2PO_4^-$, $HS^-$, $HSO_3^-$, $NCO^-$, $SCN^-$, $SO_3^{2-}$, $SO_4^{2-}$, $HPO_4^{2-}$, $CO_3^{2-}$). The mechanism was investigated by absorption and emission spectra together with $^1$H-NMR titration.

## 1. Introduction

In the past decades, the studies on designing and synthesizing new chemical sensors for detecting anions had been an remarkable topic because of their key functions in biological, chemical and environmental systems[1,2]. Cyanide ($CN^-$), a highly toxic anion, has drawn special interest due to its affection to many biological functions and widely applications in numerous chemical processes, such as electroplating, plastic manufacturing, gold extraction, tanning and metallurgy[3]. Many solutions have been proposed to detect cyanide, for example, titrimetric, electrochemical, and chromatography[4-7]. Among these solutions, optical sensors have drawn much attention, as their higher selectivity, sensitivity and low cost[8.9]. Hence, several chemosensor systems for $CN^-$ detection have been developed, including nucleophilic addition reaction[10-12], hydrogen bonding interaction[13,14], supramolecular self-assembly[15,16], deprotonation[17], and so on. Among these strategies, sensors based on nucleophilic addition reaction showed both specific selectivity and high sensitivity, due to the selective reaction with cyanide anion.

In this work, we synthesized a new highly selective 'turn-on' fluorescent and colorimetric chemosensor, 2-((4,7-bis(4-(diphenylamino)phenyl)benzo[c][1,2,5]thiadiazol-5-yl)methylene)malononitrile (DTPA-BT), for cyanide detection which was based on triphenylamine and benzothiadiazole. In the molecular structure, the triphenylamine-benzothiadiazole unit displays excellent potential to be an optical platform for detecting, because it can be modulated easily and variously. Moreover this sensor has a large stokes shift, as a result, the fluorescence wavelength of DTPA-BT is near infrared, which is helpful for biosensing. In our strategy, the dicyanovinyl group was utilized as the sensing unit to realize the nucleophilic - addition reaction. The $CN^-$ (nucleophilic ion) can be bonded with dicyanovinyl group resulting from the electron-withdrawing feature, which breaks conjugation through nucleophilic addition reaction. Upon the addition of $CN^-$, the spectra changed rapidly, and showed high sensitivity. Furthermore, we investigated the response towards $CN^-$ over other anions $F^-$, $Cl^-$, $Br^-$, $I^-$, $NO_2^-$, $NO_3^-$, $HCO_3^-$, $H_2PO_4^-$, $HS^-$, $HSO_3^-$, $NCO^-$, $SCN^-$, $SO_3^{2-}$, $SO_4^{2-}$, $HPO_4^{2-}$, $CO_3^{2-}$ and found that it performances well both in selectivity and anti-interference.

## 2. Results and discussion

### *2.1 Design and synthesis of sensor DTPA-BT*

The structure of DTPA-BT contained both triphenylamine and benzothiadiazole moieties, in which a dicyanovinyl group was attached to act as a sensing unit (**Scheme 1**). Moreover, the electrophilic nature of the dicyanovinyl substituent can be modulated by $CN^-$, which interrupts the $\pi$-conjugation[18].

DTPA-BT was prepared by Knoevenagel condensation reaction[19], in 92% yield. The synthetic routes of DTPA-BT were shown in Scheme 1. Detailed synthetic procedures and product characterizations are provided in the Experimental section and Supplementary data (**Fig.S1-S18**).

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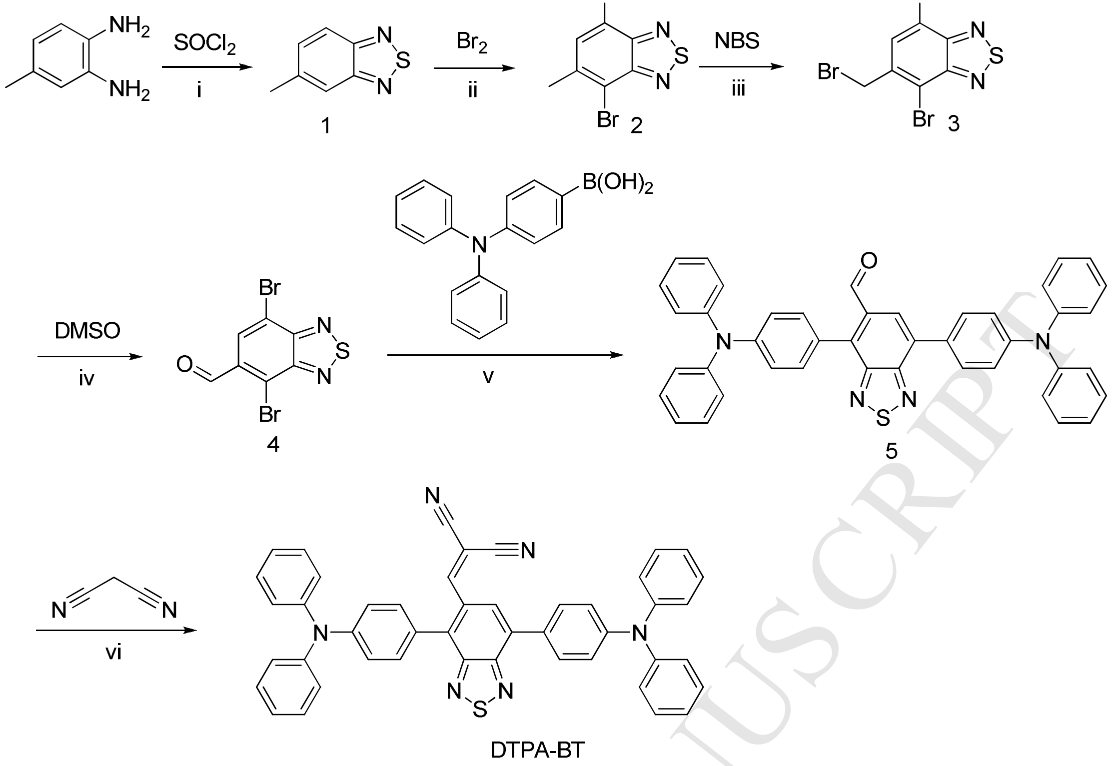

(i) $CH_2Cl_2$/TEA, reflux, 5h, 82%. (ii) HBr (47%), reflux, 6h, 71%. (iii) $CCl_4$, reflux, 17h, 80%. (iv) reflux, 0.5h, 76%. (v) THF/$H_2O$/$K_2CO_3$, reflux, 12h, 56%. (vi) AcOH/$AcONH_4$, reflux, 8h, 92%.

**Scheme 1.** Synthetic route of compound DTPA-BT

### *2.2 Absorption spectral response to cyanide ions*

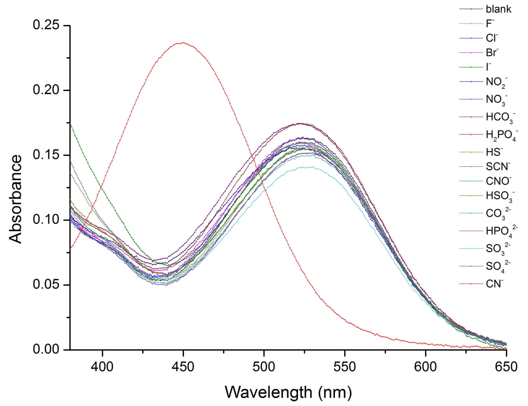

**Fig.1.** Absorption spectra of DTPA-BT ($1\times10^{-5}$ M) in a THF/H2O (99:1 [v/v]) solution upon addition of 2.0 equiv. of various anions.

The sensing properties of DTPA-BT were examined in THF/$H_2O$ solutions (99:1 [v/v]) through the addition of sodium salt of various anions, including $F^-$, $Cl^-$, $Br^-$, $I^-$, $NO_2^-$, $NO_3^-$, $HCO_3^-$, $H_2PO_4^-$, $HS^-$, $HSO_3^-$, $NCO^-$, $SCN^-$, $SO_3^{2-}$, $SO_4^{2-}$, $HPO_4^{2-}$, $CO_3^{2-}$. Upon the addition of 2.0 equivalent (equiv.) of various anions (except for the $CN^-$ ion), the absorption spectra of DTPA-BT did not show any substantial change (**Fig. 1**). However, upon the gradual addition of $CN^-$ ion, the absorption band centered at 521 nm was apparently decreased, which was attributed to the intramolecular charge transfer (ICT) transition in the molecule, and a new band centered at 450 nm was generated. The color of the solution changed from purple to yellow, which was easily observed by the naked eyes (**Fig. 5(a)**). A gradual spectral change was also shown in **Fig. 2**, where a clear isosbestic point can be observed at 487 nm, indicating a clear transformation to a new species. The Fluorescence quantum yield of DTPA-BT with $CN^-$ (2 equiv.) is 0.23.

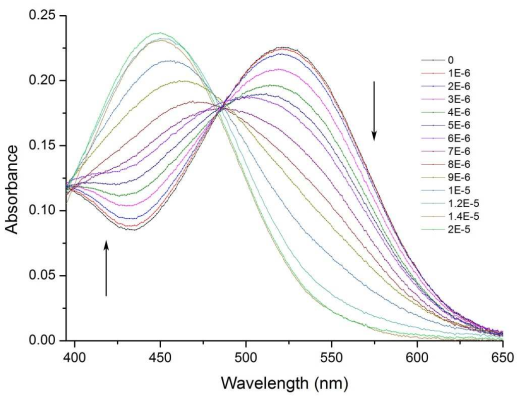

**Fig.2.** Absorption spectra of DTPA-BT ($1\times10^{-5}$ M) in a THF/$H_2O$ (99:1 [v/v]) solution upon gradual addition of increasing amounts of cyanide anions (from 0 to 2.0 equiv.).

### *2.3 Anions-induced fluorescence spectral response to cyanide ions*

The excitation of the sensor DTPA-BT at 480 nm produced an emission peak at 627 nm, showing a marvelous stokes shift nearly 150 nm, and gave an orange-red emission. Upon the addition of $F^-$, $Cl^-$, $Br^-$, $I^-$, $NO_2^-$, $NO_3^-$, $HCO_3^-$, $H_2PO_4^-$, $HS^-$, $HSO_3^-$, $NCO^-$, $SCN^-$, $SO_3^{2-}$, $SO_4^{2-}$, $HPO_4^{2-}$, $CO_3^{2-}$ no significant change was observed in the emission spectra of DTPA-BT (**Fig. 3**). The only substantial response appeared when $CN^-$ was added, and the band at 627 nm remarkably increased. This color change of the solution was easily observed by the naked eyes under the irradiation of a UV-lamp, which was shown in **Fig. 5(b)**. After the addition of 1.2 equiv. of $CN^-$, the peak at 627 nm was increased completely (**Fig. 4**). The fluorescent intensity (627 nm) increased linearly as a function of concentration of $CN^-$, and thus can be potentially used for quantification of $CN^-$ (**Fig. S19**). The detection limit of the fluorescent spectrum changes calculated on the basis of 3σ/K is 0.014 μM for $CN^-$, which is comparable or slightly lower than other dicanovinyl-based chemosensors[12,20].

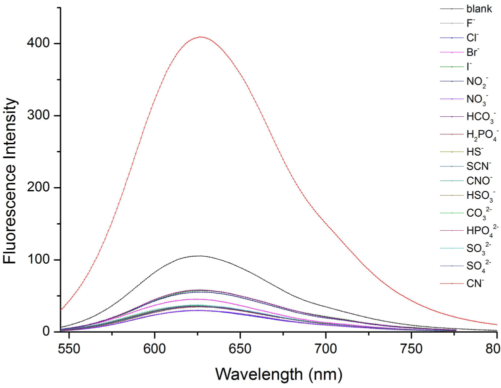

**Fig.3.** Emission spectra ($\lambda_{ex}$=480 nm) of DTPA-BT ($1\times10^{-5}$ M) in a THF/$H_2O$ (99:1 [v/v]) solution upon addition of 2.0 equiv. of various anions.

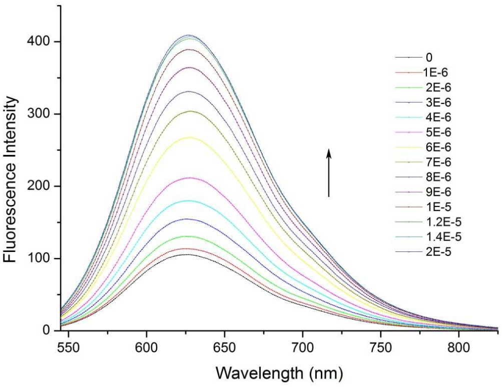

**Fig.4.** Emission spectra of DTPA-BT ($1\times10^{-5}$ M) in a THF/$H_2O$ (99:1 [v/v]) solution upon gradual addition of increasing amounts of cyanide anions (from 0 to 2.0 equiv.).

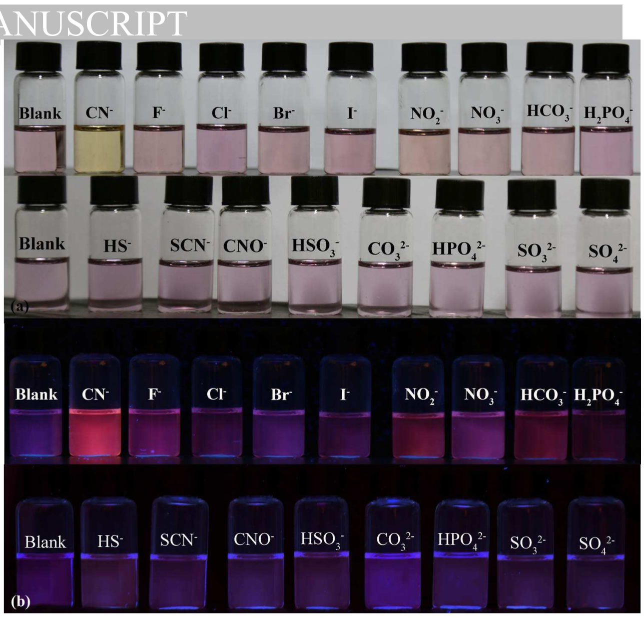

**Fig.5(a).** Naked-eye visible images of DTPA-BT ($1\times10^{-5}$ M) upon the addition of 2.0 equiv. of $CN^-$ and other interfering anions in a THF/$H_2O$ (99:1 [v/v]) solution at 25°C.

**(b).** Naked-eye fluorescent ($\lambda_{ex}$=480 nm) images of DTPA-BT ($1\times10^{-5}$ M) upon the addition of 2.0 equiv. of $CN^-$ and other interfering anions in a THF/$H_2O$ (99:1 [v/v]) solution at 25°C.

### *2.4 Selectivity and competivity investigation*

The spectra of both absorption (**Fig. 1**) and emission (**Fig. 3**), mentioned above, showed that the sensor DTPA-BT was not substantially influenced by the subsequent addition of competing anions. To evaluate the anti-interference performance of DTPA-BT, competitive experiments (**Fig. 6**) were conducted with addition of 2.0 equiv. of various other anions ($F^-$, $Cl^-$, $Br^-$, $I^-$, $NO_2^-$, $NO_3^-$, $HCO_3^-$, $H_2PO_4^-$, $HS^-$, $HSO_3^-$, $NCO^-$, $SCN^-$, $SO_3^{2-}$, $SO_4^{2-}$, $HPO_4^{2-}$, $CO_3^{2-}$) in a solution of THF/$H_2O$ (99:1 [v/v]). As shown in **Fig.6**, the fluorescence was changed prominently upon the following addition of $CN^-$ when other anions were in presence. This result confirmed the excellent selectivity and anti-interference of the dicyanovinyl-based sensor DTPA-BT for $CN^-$.

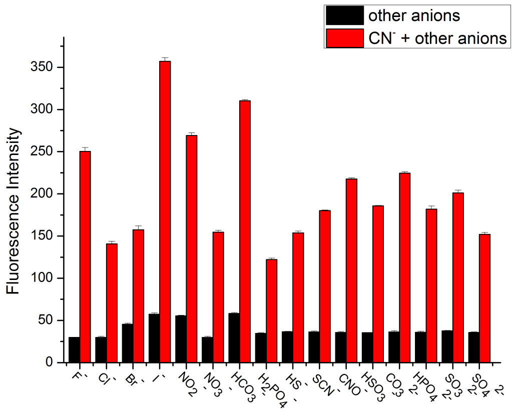

**Fig.6.** The fluorescence responses of DTPA-BT ($1\times10^{-5}$ M) upon the addition of 2.0 equiv. of $CN^-$ and other interfering anions in a THF/$H_2O$ (99:1 [v/v]) solution at 25°C (DTPA-BT + other anions, black bar; DTPA-BT + other anions + $CN^-$, red bar).

*2.5 Sensing mechanism of sensor DTPA-BT to cyanide*

The mechanism of DTPA-BT for CN$^-$ sensing was further analyzed accordingly with $^1$H-NMR spectroscopy. **Fig. 7** shows the $^1$H-NMR of DTPA-BT upon the addition of tetrabutylammonium cyanide in a solution (CDCl$_3$/DMSO-d$_6$ 99:1 [v/v]). The addition of cyanide resulted in a slow reduction of the vinylic proton signal at 8.38 ppm (H$_c$), which was disappeared finally with the addition of 1.0 equiv. of CN$^-$, while a new signal increased at 4.70 ppm (H$_c$'), corresponding to the β-proton of dicyanovinyl. Also the original aromatic protons showed a downfield shift, which was in accordance with previous studies that CN$^-$ links up the β-conjugated position of dicyanovinyl moiety[12,18,21].

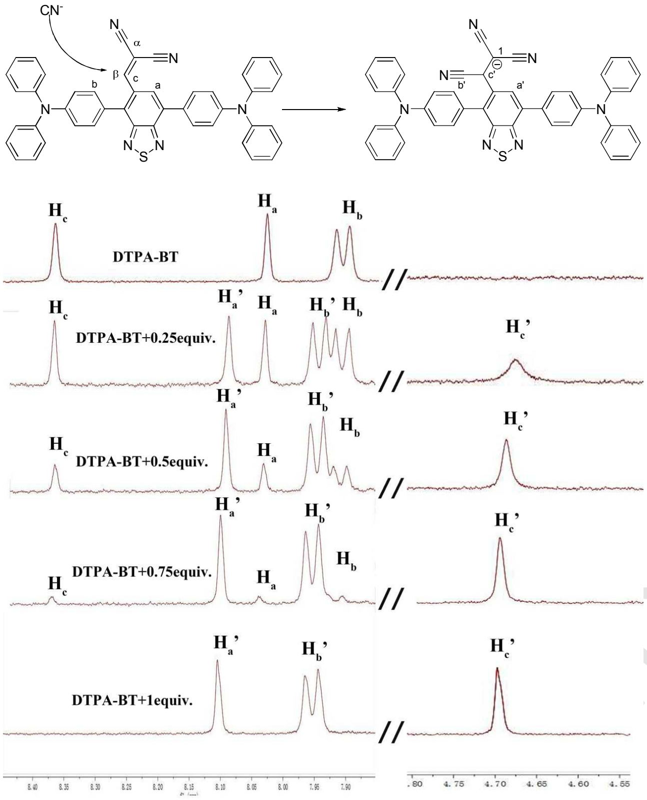

**Fig. 7.** $^1$H-NMR spectra changes of DTPA-BT in a solution (CDCl$_3$/DMSO-d$_6$ 99:1 [v/v]), upon the addition of CN$^-$ anion.

## 3. Conclusions

In summary, a colorimetric and fluorescent dual channel CN$^-$ probe (DTPA-BT), containing triphenylamine donor and benzothiadiazole acceptor was designed and synthesized. DTPA-BT could react, in principle, with 1.2 equiv. of CN$^-$. The interaction of cyanide with the probe inhibited the intramolecular charge transfer and resulted in the disappearance of the corresponding absorption peak (521 nm), and generated a new one (450 nm). Furthermore, the nucleophilic attack of CN$^-$ to the dicyanovinyl moiety induces a conspicuous emission spectra change (band centered at 627 nm increased prominently), which means it has a large stokes shift and the fluorescence wavelength is near infrared. So it has advantages to be a colorimetric and fluorescent dual channel probe. Comparing to the competitive experiments, DTPA-BT showed fast response, good selectivity and outstanding anti-interference performance.

## 4. Experimental section

*4.1 Materials and Instrumentation*

All chemical reagents and solvents were purchased from commercial suppliers and were used without further purification unless it is noted by other way. Tetrahydrofuran was dried with metal sodium and distilled immediately prior to use. All moisture-sensitive and air-sensitive reactions were carried out under argon atmosphere.

$^1$H-NMR and $^{13}$C-NMR spectra were measured on a Bruker AM-400 spectrometer using d-chloroform as a solvent and tetramethylsilane (TMS, δ = 0 ppm) as an internal standard. The UV-vis spectra were recorded on a Varian CARY 100 spectrophotometer at room temperature. Fluorescence-emission was detected on Varian Cary Eclipse spectrophotometer at room temperature in acetonitrile.

*4.2 Synthesis*

*Synthesis of 5-methylbenzo[c][1,2,5]thiadiazole* [22] **(1)**

To a 250 mL three-neck flask were added commercial 4-methylbenzene-1,2-diamine (3.75 g, 0.03 mol), 60 mL of CH$_2$Cl$_2$ and, trimethylamine (13.14 g, 0.13 mol). The solution was stirred until total dissolution of the diamine .Sulfurous dichloride (11.47 g, 0.10 mol) was added dropwise very slowly and the mixture refluxed for 5 h. After quenching the reaction with 1mol/L sodium hydroxide solution, the solvent was extracted 3 times with dichloromethane (30 mL) and removed by rotary evaporation. The residue was purified by silica gel column chromatography using pure petroleum ether as eluant to collect a brown needle solid, yield 3.69 g (82%). m.p. 33-34 °C; $^1$H-NMR (CDCl$_3$, 400 MHz, TMS) : δ 7.88 (d, 1H, *J* =9 Hz), 7.76 (s, 1H), 7.43 (d, 1H, *J* = 9 Hz), 2.55 (s, 3H); $^{13}$C-NMR (CDCl$_3$, 100 MHz, TMS) : δ 155.31, 153.54, 139.94, 132.41, 120.66, 119.76, 21.91; EI-MS [M$^+$] Calcd. 150.0252, Found 150.0251.

*Synthesis of 4,7-dibromo-5-methylbenzo[c][1,2,5]thiadiazole*[23]**(2)**

To a 250 mL three-neck flask was added 5-methyl-benzothiadiazole (6.0 g, 0.04 mol) and HBr (75 mL, 47%). A solution containing Br$_2$ (22.21 g, 0.12 mol) in HBr (10 mL) was added dropwise very slowly. After addition was complete, solution was stirred and heated under reflux for 6 h. Precipitation of a lot of solid was noted. After the reaction was cooled to room temperature, sufficient saturated solution of sodium bisulfite was added to consume excess liquid bromine. The mixture was filtrated and washed with water. The solid was then washed once with cold Et$_2$O and dried and the residue purified by silica gel column chromatography using pure petroleum ether as eluent to collect a pale yellow powder, yield 0.612 g (71%); m.p. 150-151 °C; $^1$H-NMR (CDCl$_3$, 400 MHz, TMS) : δ 7.78 (s, 1H ), 2.62 (s, 3H); $^{13}$C-NMR (CDCl$_3$, 100 MHz, TMS) : δ 153.41, 151.53, 140.43, 135.14, 114.22, 112.60, 22.67; EI-MS [M$^+$] Calcd. 305.8462, Found 305.8461.

*Synthesis of 4,7-dibromo-5-(bromomethyl)benzo[c][1,2,5]thiadiazole*[23] **(3)**

A mixture of 4,7-dibromo-5-methylbenzo[c][1,2,5]thiadiazole (1.5 g, 4.8 mmol), N-bromosuccinimide (NBS; 0.9 g, 4.8 mmol) and benzoyl peroxide (BPO; 30 mg, 0.13 mmol) was added to a 100 mL three-neck flask, which was dissolved in carbon tetrachloride (30 mL). The mixture was heated at reflux for 17 h under argon atmosphere. The solution was filtered and the filtrate concentrated by rotary evaporation. The residues were purified by silica gel column chromatography using petroleum ether-CH$_2$Cl$_2$ (4:1 (v/v)) as eluent to give a pale-yellow solid, yield 1.26 g (80%); m.p. 159-160 °C; $^1$H-NMR (CDCl$_3$, 400 MHz, TMS): δ 7.93 (s, 1H), 2.62 (s, 2H ); $^{13}$C-NMR (CDCl$_3$, 100

MHz, TMS) : δ 153.33, 152.36, 139.29, 133.97, 115.32, 113.89, 31.26; EI-MS [$M^+$] Calcd. 383.7567, Found 383.7570.

*Synthesis of 4,7-dibromobenzo[c][1,2,5]thiadiazole-5-carbaldehyde*[24,25] **(4)**

4,7-dibromo-5-(bromomethyl)benzo[c][1,2,5]thiadiazole (1.161 g, 3 mmol) and DMSO (30mL) was added to a 100 mL three-neck flask. After refluxed for 30 min, the mixture was extracted with water and dichloromethane. The solvent was removed by rotary evaporation, and the residue was purified by silica gel column chromatography using petroleum ether-$CH_2Cl_2$ (3:1 (v/v)) as eluent to give a yellow powder, yield 0.73 g (76%); m.p. > 200 °C; $^1$H-NMR ($CDCl_3$, 400 MHz, TMS): δ 10.54 (s, 1H), 8.34 (s, 1H); $^{13}$C-NMR ($CDCl_3$, 100 MHz, TMS) : δ 189.68, 154.84, 153.59, 134.08, 130.35, 121.84, 114.84. EI-MS [$M^+$] Calcd. 319.8255, Found 319.8258.

*Synthesis of 4,7-bis(4-(diphenylamino)phenyl)benzo[c][1,2,5]thiadiazole-5-carbaldehyde* **(5)**

To a mixture of 4,7-dibromobenzo[c][1,2,5]thiadiazole-5-carbaldehyde (0.22 g, 0.68 mmol) in THF (20 mL) and aqueous 2 M potassium carbonate solution (25 mL) was added 4-diphenylaminophenylboronic acid (0.8 g, 2.72 mmol) in THF (20 mL) under an argon atmosphere. Subsequently, tetrakis(triphenylphosphine)palladium (0) (78.58 mg, 0.068 mmol) was added to the mixture. After the mixture was refluxed for 12 h, the reaction mixture was poured into water and extracted with dichloromethane. The organic layer was dried over anhydrous sodium sulfate and evaporated to dryness. The residue was purified by silica gel column chromatography using petroleum ether-$CH_2Cl_2$ (4:1 (v/v)) as eluent to give a red powder, yield 0.25g (56%); m.p. > 200 °C; $^1$H-NMR ($CDCl_3$, 400 MHz, TMS): δ 10.23 (s, 1H), 8.29 (s, 1H), 7.91 (d, 2H, $J$ = 8 Hz), 7.50 (d, 2H, $J$ = 8 Hz), 7.36-7.29 (m, 8H), 7.24-7.19 (m, 12H), 7.14-7.06 (m, 4H); $^{13}$C-NMR ($CDCl_3$, 100 MHz, TMS): δ 192.30, 155.84, 155.64, 149.17, 148.47, 147.38, 147.04, 138.68, 133.07, 132.77, 130.00, 129.57, 129.42, 125.57, 125.05, 124.50, 124.06, 123.49, 122.65, 121.13; ESI-MS [$M+Na^+$] Calcd. 673.2033, Found 673.2042.

*Synthesis of 2-((4,7-bis(4-(diphenylamino)phenyl)benzo[c][1,2,5]thiadiazol-5-yl)methylene)malononitrile* **(DTPA-BT)**

A mixture of 4,7-bis(4-(diphenylamino)phenyl)benzo[c][1,2,5]thiadiazole-5-carbaldehyde (0.065 g, 0.1 mmol), ammonium acetate (0.154 g, 2 mmol), glacial acetic acid (80 mL) and malononitrile (0.066 g, 1 mmol) was refluxed under an argon atmosphere for 8 h. After cooling to room temperature, the mixture was poured into water (300 mL), and then extracted with dichloromethane (50 mL×3). The organic layer was concentrated and residue was purified by silica gel column chromatography using petroleum ether-$CH_2Cl_2$ (1:1 (v/v)) as eluent to give a black purple powder, yield 0.064 g (92%); m.p. > 200 °C; $^1$H-NMR (THF-$d_8$, 400 MHz, TMS): δ 8.44 (s, 1H), 8.22 (s, 1H), 8.03 (d, 2H, $J$ = 8 Hz), 7.50 (d, 2H, $J$ = 8 Hz), 7.35-7.28 (m, 8H), 7.23-7.16 (m, 12H), 7.13-7.01 (m, 4H); $^{13}$C-NMR( $CDCl_3$, 100 MHz, TMS): δ 158.28, 154.05, 153.87, 148.75, 147.81, 146.19, 145.58, 138.20, 132.17, 131.81, 129.06, 128.66, 128.41, 125.07, 124.23, 123.58, 122.66, 121.34, 119.08, 113.05; EI-MS [$M^+$] Calcd. 698.2253 , Found 698.2252

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## Supplementary data

$^1$H-NMR, $^{13}$C-NMR, and MS of DTPA-BT are available, and additional graphs for calculation of detection limit can be found in ESI.

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