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# Dual responsive chemosensors for anions: the combination of fluorescent PET (Photoinduced Electron Transfer) and colorimetric chemosensors in a single molecule

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**Abstract**—The design and synthesis of two novel fluorescent PET anion sensors is described, based on the principle of '*fluorophore-spacer-(anion)receptor*'. The sensors **1** and **2** employ simple diaromatic thioureas as anion receptors, and the fluorophore is a naphthalimide moiety that absorbs in the visible part of the spectrum and emits in the green. Upon recognition of anions such as $F^-$ and $AcO^-$ in DMSO, the fluorescence emission of **1** and **2** was '*switched off*', with no significant changes in the UV–vis spectra. This recognition shows a 1:1 binding between the receptor and the anions. In the case of $F^-$, further additions of the anion, gave rise to large changes in the UV–vis spectra, where the $\lambda_{max}$ at 455 nm was shifted to 550 nm. These changes are thought to be due to the deprotonation of the 4-amino moiety of the naphthalimide fluorophore. This was in fact found to be the case, using simple naphthalimide derivatives such as **6**. Sensors **1** and **2** can thus display dual sensing action; where at low concentrations, the fluorescence emission is quenched, and at higher concentrations the absorption spectra are modulated.


There is considerable interest within the field of supramolecular chemistry in the design of chemosensors for cation and anion recognition.[1] In this regard several excellent examples of fluorescent and colorimetric sensors for cations have been developed.[1,2] However, the area of anion recognition and sensing is much less explored.[3,4] We are particularly interested in anion sensing and have designed several Tb(III) cyclen complexes as chemosensors for aromatic carboxylates such as salicylic acid.[5] We recently also developed the first examples of fluorescent anion PET chemosensors using simple charge neutral receptors.[6] Here the recognition event occurred through hydrogen bonding between urea or thiourea receptors and anions with concomitant quenching of fluorescence via a PET mechanism.[7] Unfortunately, a drawback to this design was the short emission wavelength of the fluorophore (anthracene), which suffers from the affects of background autofluorescence and light scattering.[8] Accordingly, we set out to design new PET chemosensors that emit at longer wavelengths and chose 4-amino-1,8-naphthalimide as a fluorophore, as it emits in the green ($\lambda_{max} \sim$ 540–550 nm) with high quantum yields ($\Phi_F$).[9] Moreover, we aimed to improve our receptor design by incorporating a second aromatic unit within the thiourea scaffold with the aim of further enhancing the sensitivity of anion recognition. Compounds **1** and **2** best exemplify our current design strategy. Additionally, due to the presence of a large dipole moment in the naphthalimide, which originates from an internal charge transfer excited state (ICT),[10] the ground state, and therefore the color of the compounds, may be modulated via hydrogen bonding or deprotonation of the 4-amino moiety by small anions possessing high charge density such as $F^-$ and $HO^-$. The dual action of **1** and **2** would thus yield a combined *fluorescence* and *colorimetric* based sensor in a *single molecule*. To the best of our knowledge, this would be the first such example of a combined fluorescent and colorimetric system.

The two chemosensors are designed on the classic *fluorophore-spacer-receptor* principle developed by de Silva et. al.[11] The synthesis of **1** and **2** is shown in Scheme 1. Both sensors are made from readily available starting materials. The precursor to both was **4**, made in two steps in 90% yield form 4-bromo-1,8-naphthalic

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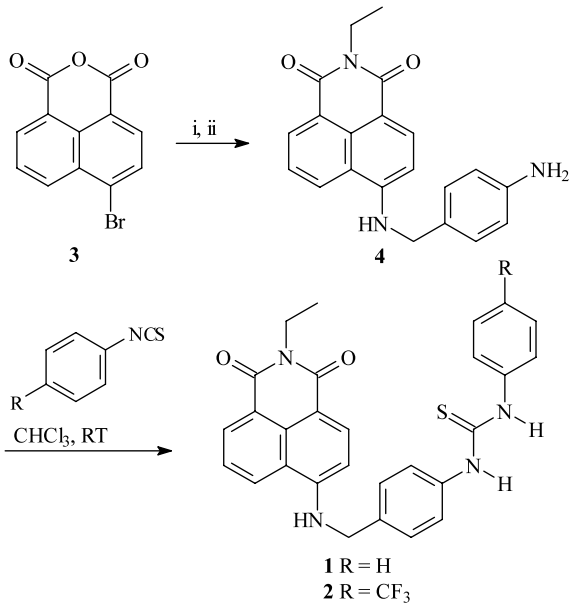



**Scheme 1.** The synthesis of **1** and **2** from the starting material **3** in two steps: (i) $CH_3CH_2NH_2$, 1,4-dioxane, reflux; (ii) 4-aminobenzylamine (neat) 130°C.

anhydride **3**, by first reacting it with ethylamine (70%) in 1,4-dioxane; the desired product being precipitated upon addition to water. This was followed by the nucleophilic displacement using an excess of 4-aminobenzylamine (neat) at 130° C, followed by recrystallization from ethanol. Consequently, the two anion sensors were formed by reacting **4** with phenyl- or 4-(trifluoromethyl)phenyl isothiocyanate at room temperature in dry $CHCl_3$ giving **1** and **2** in 60 and 58% yields, respectively (Scheme 1). All intermediates and sensors were characterized using conventional methods (See Reference Section). Both sensors showed the presence of two thiourea protons in their $^1$H NMR spectra in DMSO-$d_6$. For **1** these appeared as broad resonance at 9.72 ppm, whereas the amino proton appeared as a broad singlet at 8.43 ppm. Similar results were observed for **2**, with resonances at 10.02 ppm and 8.45 ppm, respectively.

The ability of **1** and **2** to recognize several anions was investigated using fluorescence and UV–vis spectroscopy in DMSO. Both sensors were highly colored with $\lambda_{max}$ at ca. 444 nm ($\log \varepsilon \sim 4.20$), which was assigned to the ICT excited state. Excitation at $\lambda_{max}$ gave rise to strong emission between 450 and 700 nm (green to the naked eye) with $\Phi_F = 0.60$ for **1** and 0.71 for **2**. Upon addition of several anions such as $AcO^-$, $H_2PO_4^-$ and $F^-$ the emission of **1** and **2** ($\sim$6 mM) was substantially reduced in intensity. The mechanism for this quenching is via PET, which takes place between the receptor and the fluorophore. Unlike many PET sensors for cations, the fluorescence of **1** and **2** is '*switched off*' rather then '*switched on*' upon ion recognition. We propose that this quenching process is due to the following; prior to the recognition process, the excited state of the fluorophore is not, or only to a

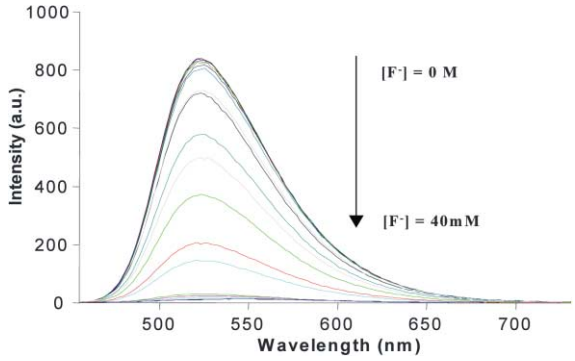



**Figure 1.** The changes in the fluorescence emission spectra of **1** (6 μM) upon titration with solutions of $F^-N(C_4H_9)_4^+$ in DMSO, showing the fluorescence emission being '*switched off*' upon $F^-$ recognition. All measurements were repeated several times.

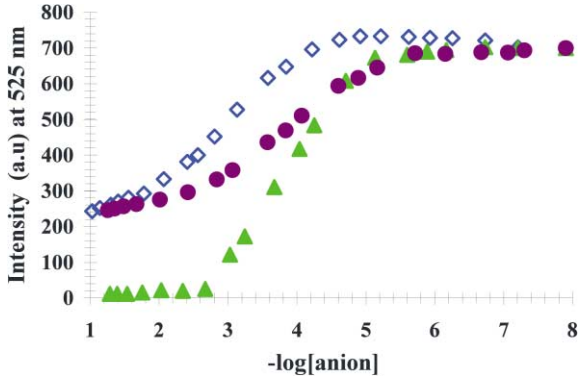



**Figure 2.** Changes in the emission spectra of **1** at 525 nm as a function of –log[anion]: $F^-$ (green triangle); $AcO^-$ (purple circle); $H_2PO_4^-$ (blue diamond).

minor extent, quenched by electron transfer from the receptor to the fluorophore. However, after the addition of the anion, and the formation of the *anion-receptor hydrogen bonding complex*, the reduction potential of the receptor is increased, making the electron transfer more feasible. This subsequently gives rise to enhanced fluorescence quenching. This is clearly demonstrated in Figure 1 for the addition of $F^-$ to **1** in DMSO. Here the fluorescence emission of **1** is effectively quenched or completely '*switched off*' after the addition of 40 mM of $F^-$. Concurrently, only minor changes were seen in the absorption spectra of **1** at this concentration of $F^-$ (see later). Addition of MeOH (ca. 10% v/v) to this solution '*re-switched on*' the emission, demonstrating that the process was fully reversible, i.e. the hydrogen bonding interactions were broken.

Similar effects were observed in the emission spectra of **1** upon addition of $AcO^-$ and $H_2PO_4^-$, but the quenching factors were somewhat smaller. The addition of $Cl^-$ and $Br^-$ did not lead to any significant changes in the emission of **1**, possibly due to their large size and low charge density. Plotting the changes at 525 nm ($\lambda_{Fmax}$) as a function of –log [anion] gave in all cases, sigmoidal curves that '*switched off*' over two log units (Fig. 2).

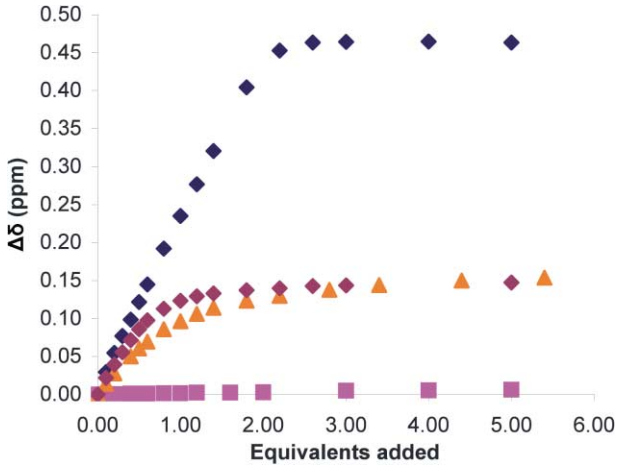



**Figure 3.** The changes $^1$H NMR (400 MHz, DMSO-$d_6$) in one of the aromatic protons of **1** upon titration of several anions: $F^-$ (blue diamond), $AcO^-$ (red diamond); $H_2PO_4^-$ (orange triangle); $Cl^-$ (pink square).

These changes were fitted using a non-linear least squares regression algorithm, giving the binding constants log $\beta = 3.8(\pm 0.1)$, $3.9(\pm 0.1)$ and $2.9(\pm 0.1)$ for $F^-$, $AcO^-$ and $H_2PO_4^-$ respectively. Even though the binding of $F^-$ and $AcO^-$ was quite similar more effective quenching was seen for the former. We attribute this to stronger hydrogen bonding between the $F^-$ and the thiourea protons, since $F^-$ is smaller and has higher charge density then $AcO^-$. In the case of **2**, the rather simple modification of the receptor site, the presence of the aryl-$CF_3$ group, further increased the acidity of the thiourea hydrogens giving rise to slightly stronger binding with log $\beta = 4.4$ $(\pm 0.1)$ and 3.7 $(\pm 0.1)$ for $F^-$ and $H_2PO_4^-$ and 4.0 $(\pm 0.2)$ for $AcO^-$.[†] We have recently developed several families of such bis-aromatic ureas and thioureas as anion receptors, and measured their binding interactions with several 'simple' anions and amino acids using $^1$H NMR. These clearly show that the binding affinity and the selectivity of these kinds of receptors can be tuned.[12]

These binding interactions were also monitored using $^1$H NMR in DMSO-$d_6$, by monitoring the aromatic protons as the thiourea protons became too broad upon addition of $F^-$, $AcO^-$ and $H_2PO_4^-$. For $AcO^-$ and $H_2PO_4^-$ the plot of $\Delta\delta$ versus equivalents added indicated 1:1 binding by **1** and **2**, whereas for $F^-$ 1:2 binding was determined for both sensors. No binding was seen for $Cl^-$ or $Br^-$. These results are shown in Figure 3. The 1:2 binding was also visible to the naked eye after the addition of ca. 2–2.5 equiv. of $F^-$ as the color of the solution changed from light yellow to deep purple. These color changes signify the interaction of $F^-$ with the 4-amino moiety which is either through strong hydrogen bonding between N–H···F, or more likely by complete deprotonation. This significantly

[†] In the case of $AcO^-$, the emission was quenched, and log $\beta$ reported was determined from those changes. However, at higher concentrations, the emission was enhanced again. We did not see this effect for any of the other anions. We are currently investigating this in greater detail.

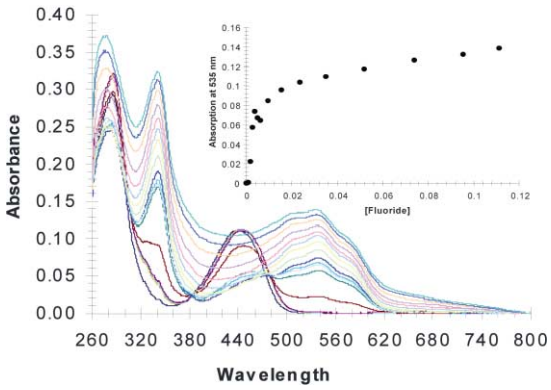
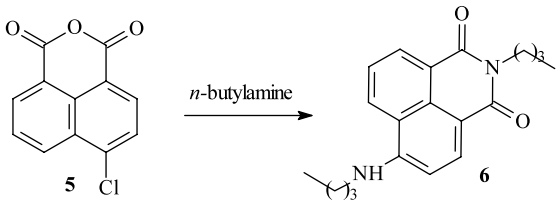



**Figure 4.** The changes in the absorption spectra (uncorrected) of **1** upon addition of $F^-$. Insert: the changes at 525 nm as a function of $[F^-]$.




**Scheme 2.** The one-pot synthesis of **6** from the anhydride **5**.

increases the charge density on the amino nitrogen with concurrent enhancement in the push–pull character of the ICT.

The concomitant changes in the UV–vis spectra for **1** upon addition of $F^-$ can be seen in Figure 4. As stated previously no major changes were seen with up to 30 mM of $F^-$. However, upon further addition, the absorption spectra were dramatically affected. In the first instance, the absorption at 444 nm was reduced with the formation of a new band centered at 536 nm, tailing to 620 nm, with an isosbestic point at 475 nm. Upon further increasing the concentration the band at ca. 410 nm also increased, which is possibly due to aggregation. Similar results were observed using tetrabutyl ammonium hydroxide, indicating that deprotonation had indeed occurred. Upon excitation of the newly formed 535 nm band, a new weak and broad fluorescence emission was observed between 580 and 800 nm, which was $[F^-]$ dependent. Addition of MeOH reversed these changes. Similar results were also seen for **2**, indicating that **1** and **2** can both detect $F^-$ at two different $[F^-]$ ranges with two different sensing modes. To the best of our knowledge these are the first examples of such dual *fluorescent-colorimetric sensors* for anions, e.g. the naphthalimide emission is quenched by the anion recognition at the thiourea receptor site, whereas hydrogen bonding/deprotonation at the 4-amino moiety gave rise to large color changes resulting in the formation of dual responsive chemosensors for anions such as $F^-$.

To investigate this phenomena further, we synthesized the reference compound **6**, by reacting in a one-pot synthesis the starting material 4-chloro-1,8-naphthalic

anhydride, **5**, in neat *n*-butylamine at 80°C (Scheme 2).[9] The resulting solution was then poured over ice-water to give **6** as a yellow solid in 72% yield. The addition of $F^-$ to a solution of **6** in DMSO shifted the absorption spectra of **6** to the red, in a similar manner as shown for **1** above. These changes mirrored that of **1** at high $F^-$ concentrations, but no PET quenching was observed since the molecule lacks the thiourea receptor. We are currently investigating molecules such as **6** to act as anion (pH) sensors in organic solutions.[12]

In summary, we have developed new, combined PET and colorimetric chemosensors for anions, which give rise to large changes in the fluorescence at long wavelengths, and naked eye detectable color changes from green to purple for **1** and **2** within two concentration ranges. We are currently modifying the design of **1** and **2** with the aim of enhancing the use of such fluorescent–colorimetric dual anion sensors.

## Acknowledgements

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## References

1. (a) Kimura, E. *Acc. Chem. Res.* **2001**, *34*, 171; (b) Kaminskaia, N. V.; Spingler, B.; Lippard, S. J. *J. Am. Chem. Soc.* **2001**, *123*, 7831; (c) Rurack, K.; Resch-Genger, U. *Chem. Soc. Rev.* **2002**, 116; (d) Lavigne, J. J.; Anslyn, E. V. *Angew. Chem., Int. Ed.* **2001**, *40*, 3119; (e) deSilva, A. P.; Fox, D. B.; Huxley, A. J. M.; Moody, T. S. *Coord. Chem. Rev.* **2000**, *205*, 41; (f) Fabbrizzi, L.; Licchelli, M.; Rabaioli, G.; Taglietti, A. F. *Coord. Chem. Rev.* **2000**, *205*, 85; (g) deSilva, A. P.; Fox, D. B.; Huxley, A. J. M.; McClenaghan, N. D.; Roiron, J. *Coord. Chem. Rev.* **1999**, *186*, 297.
2. (a) Amendola, V.; Fabbrizzi, L.; Mangano, C.; Pallavicini, P. *Acc. Chem. Res.* **2001**, *34*, 488; (b) Czarnik, A. W. *Acc. Chem. Res.* **1994**, *27*, 302; (c) Gunnlaugsson, T.; Leonard, J. P. *J. Chem. Soc., Perkin Trans. 2* **2002**, 1980; (d) Gunnlaugsson, T.; Nieuwenhuyzen, M.; Richard, L.; Thoss, V. *J. Chem. Soc., Perkin Trans. 2* **2002**, 141.
3. (a) Gale, P. A. *Coord. Chem. Rev.* **2001**, *213*, 79; (b) Gale, P. A. *Coord. Chem. Rev.* **2000**, *199*, 181; (c) Beer, P. D.; Gale, P. A. *Angew. Chem., Int. Ed.* **2001**, *40*, 486; (d) Beer, P. D. *Chem. Commun.* **1996**, 689.
4. (a) Moshihisa, M.; Wei, W.-H.; Eller, L. R.; Sessler, J. L. *J. Am. Chem. Soc.* **2002**, *124*, 1134; (b) Liao, J. H.; Chen, C. T.; Fang, J. M. *Org. Lett.* **2002**, *4*, 561; (c) Jiménez, D.; Martínez-Máñez; Sancenòn, F.; Soto, J. *Tetrahedron Lett.* **2002**, *43*, 2823; (d) Lee, D. H.; Lee, K. H.; Hong, J. I. *Org. Lett.* **2001**, *3*, 5; (e) Kruger, P. E.; Mackie, P. R.; Nieuwenhuyzen, M. *J. Chem. Soc., Perkin Trans. 2* **2001**, 1079; (f) Wiskur, S. L.; Best, M. D.; Lavigne, J. J.; Schneider, S. E.; Perreault, D. M.; Monahan, M. K.; Anslyn, E. V. *J. Chem. Soc., Perkin Trans. 2* **2001**, 315; (g) Cooper, C. R.; Spencer, N.; James, T. D. *Chem. Commun.* **1998**, 1365.
5. Gunnlaugsson, T.; Harte, A. J.; Leonard, J. P.; Nieuwenhuyzen, M. *Chem. Commun.* **2002**, 2134.
6. Gunnlaugsson, T.; Davis, A. P.; Glynn, M. *Chem. Commun.* **2001**, 2556.
7. Gunnlaugsson, T.; Davis, A. P.; O'Brien, J. E.; Glynn, M. *Org. Lett.* **2002**, *4*, 2449.
8. (a) Gunnlaugsson, T.; MacDónaill, D. A.; Parker, D. *J. Am. Chem. Soc.* **2001**, *123*, 12866; (b) Gunnlaugsson, T. *Tetrahedron Lett.* **2001**, *42*, 8901; (c) Gunnlaugsson, T.; MacDónaill, D. A.; Parker, D. *Chem. Commun.* **2000**, 93; (d) Gunnlaugsson, T.; Parker, D. *Chem. Commun* **1998**, 511.
9. (a) de Silva, A. P.; Rice, T. E. *Chem. Commun.* **1999**, 163; (b) de Silva, A. P.; Gunaratne, H. Q. N.; Gunnlaugsson, T. *Tetrahedron Lett.* **1998**, *39*, 5077.
10. de Silva, A. P.; Gunaratne, H. Q. N.; Habib-Jiwan, J.-L.; McCoy, C. P.; Rice, T. E.; Soumillion, J.-P. *Angew. Chem., Int. Ed. Engl.* **1995**, *34*, 1728.
11. deSilva, A. P.; Gunaratne, H. Q. N.; Gunnlaugsson, T.; Huxley, A. J. M.; McCoy, C. P.; Rademacher, J. T.; Rice, T. E. *Chem. Rev.* **1997**, *97*, 1515.
12. Manuscript in preparation.
**Synthesis**:
**4** was obtained as light fluffy yellow needles (1.53 g, 90%). $^1$H NMR (400 MHz, DMSO-$d_6$): 1.17 (t, 3H, $J=7.0$ Hz), 4.04 (q, 2H, $J=7.0$ Hz), 4.54 (d, 2H, $J=5.2$ Hz) 4.95 (bs, -$NH_2$), 6.54 (d, 2H, $J=8.0$ Hz), 6.70 (d, 1H, $J=8.54$ Hz), 7.07 (d, 2H, $J=8.0$ Hz), 7.68 (t, 1H, $J=7.5$ Hz), 8.18 (d, 1H, $J=8.5$), 8.42 (bs, -NH), 8.43 (d, 1H, $J=8.5$ Hz), 8.74 (d, 1H, $J=8.5$ Hz); $^{13}$C NMR (400 MHz, DMSO-$d_6$); $^{13}$C NMR (400 MHz, DMSO-$d_6$): 163.53, 162.67, 150.54, 147.70, 133.92, 130.54, 129.37, 128.52, 127.93, 124.93, 124.27, 121.95, 120.26, 113.93, 107.81, 104.51, 45.86, 34.23, 13.29; $m/z$ (ESMS) 346 (M+H$^+$). Anal. calcd for $C_{21}H_{19}N_3O_2$: C, 73.03; H, 5.54; N, 12.17. Found: C, 72.76; H, 5.63; N, 12.06.
**1** was obtained as a light yellow solid (0.191 g, 58%). $^1$H NMR (400 MHz, DMSO-$d_6$): 1.17 (t, 3H, $J=7.0$ Hz), 4.05 (q, 2H, $J=7.0$ Hz), 4.64 (d, 2H, $J=5.2$ Hz), 6.71 (d, 2H, $J=8.5$ Hz), 7.11 (t, 1H, $J=7.5$), 7.29–7.38 (m, 4H), 7.44–7.48 (m, 4H), 7.72 (t, 1H, $J=7.5$), 8.20 (d, 1H, $J=8.5$), 8.43 (bs, -NH), 8.47 (d, 1H, $J=8.5$ Hz), 8.78 (d, 1H, $J=8.5$ Hz), 9.72 (bs, -NH); $^{13}$C NMR (100 MHz, DMSO-$d_6$): 179.62, 163.53, 162.70, 150.38, 143.41, 138.08, 134.90, 133.92, 130.67, 129.38, 128.49, 127.14, 125.56, 125.52, 124.51, 123.93, 122.75, 122.06, 120.33, 113.02, 108.28, 104.58, 45.57, 34.25; $m/z$ 481 (M+H$^+$). Anal. calcd for $C_{28}H_{24}N_4O_2S.CH_2Cl_2.CHCl_3$: C, 52.61; H, 3.97; N, 8.18. Found: C, 52.59; H, 4.00; N, 8.39.
**2** was obtained as a light yellow solid (0.245 g, 60%). $^1$H NMR (400 MHz, DMSO-$d_6$): 1.18 (t, 3H, $J=7.0$ Hz), 4.05 (q, 2H, $J=7.0$ Hz), 4.66 (d, 2H, $J=5.2$ Hz), 6.72 (d, 1H, $J=8.5$ Hz), 7.38–7.47 (m, 4H), 7.65–7.76 (m, 4H), 8.29 (d, 1H, $J=8.5$), 8.45 (bs, NH), 8.47 (d, 2H, $J=7.5$ Hz), 8.74 (d, 1H, $J=8.5$ Hz), 10.02 (bd, -NH). $^{13}$C NMR (100 MHz, DMSO-$d_6$): 179.62, 163.53, 162.70, 150.38, 143.41, 138.08, 134.90, 133.92, 130.67, 129.38, 128.49, 127.14, 125.56, 125.52, 124.51, 123.93, 122.75, 122.06, 120.33, 113.02, 108.28, 104.58, 45.57, 34.25, 13.30; $m/z$ (ESMS) 549 (M+H$^+$). Anal. calcd for $C_{29}H_{23}F_3N_4O_2S$ $CHCl_3$: C, 53.94; H, 3.62; N, 8.39. Found: C, 53.89; H, 3.83; N, 8.40.