Synthesis of ferrocenyl- and pyrenyl-thioimidates of terminal acetylenes. “Click” reaction with 3′-azido-3′-deoxythymidine affording redox-active and fluorescent thymidine conjugates

Anna Wrona-Piotrowicz, Damian Plażuk, Sławomir Domagała, and Janusz Zakrzewski

Department of Organic Chemistry, University of Łódź, Tamka 12, 91-403 Łódź, Poland
Department of Inorganic and Analytical Chemistry, University of Łódź, Tamka 12, 91-403 Łódź, Poland
E-mail: janzak@uni.lodz.pl

DOI: http://dx.doi.org/10.3998/ark.5550190.0013.636

Abstract
Thioamides Ar(C=S)NHCOOEt (Ar = ferrocenyl and 1-pyrenyl) obtained from ferrocene and pyrene and ethoxycarbonyl isothiocyanate react under Mitsunobu conditions with propargyl and homopropargyl alcohols, yielding the corresponding N- (ethoxycarbonyl)thioimidates containing a terminal acetylene functionality. These compounds undergo Cu(I)-catalyzed “click reaction” with 3′-azido-3′-deoxythymidine (AZT) affording novel redox-active ferrocenyl and fluorescent, environmentally sensitive pyrenyl thymidine conjugates.

Keywords: Ferrocene, pyrene, AZT, thioamide, click reactions, fluorescence

Introduction
Labeling of biomolecules with redox active or fluorescent reporter groups constitutes a powerful research tool for biological and medical sciences. Among various redox-active markers ferrocenyl compounds are of increased interest due to the accessibility of a large variety of compounds, reversibility of the redox process and the easy tuning by changing substituents at the cyclopentadienyl ligands. On the other hand, pyrene derivatives are attractive fluorescent markers due to long lifetimes of their excited states, ability to π-stacking resulting in exciplex and excimer formation and sensitivity to the polarity of the surrounding medium.

We have recently reported that ferrocenyl thioamide Fc(C=S)NHCOOEt (1a) reacts as an S-pronucleophile in the Mitsunobu reaction with alcohols to afford the corresponding ferrocenyl (N-ethoxycarbonyl)thioimidates. This reaction has been applied for redox labeling of adenosine, 2′-deoxyadenosine and some steroidal alcohols.
Herein we report that 1a can be readily transformed by Mitsunobu reaction into thioimidates 3a–b (Scheme 1) with terminal acetylene groups. These compounds can be used as “clickable” markers for azide-functionalized biomolecules as exemplified by 3’-azido-3’-deoxothymidine (AZT, 4), applying the Cu(I)-catalyzed Huisgen-Meldal azide-alkyne cycloaddition (CuAAC),\(^5\) thus affording redox-active conjugates 5a,b. Furthermore, we have extended this approach to pyrenyl compounds, synthesizing the N-ethoxycarbonyl thioamide 1b, acetylenic thioimidates 3c,d, and AZT conjugates 5c,d. We have found that these conjugates show intense solvent-dependent fluorescence.

**Results and Discussion**

It has been reported that the Friedel-Crafts reaction of ferrocene with N-ethoxycarbonyl isothiocyanate carried out in methanesulfonic acid at rt affords thioamide 1a in 48% yield.\(^6\) We have found that with pyrene this reaction proceeds better in the presence of trifluoromethanesulfonic acid in dichloromethane affording thioamide 1b in 95% yield. The \(^{13}\)C NMR signals of the thiocarbonyl and carbonyl groups of this compound appear at \(\delta\) 201.5 and 149.5, respectively. The Mitsunobu reaction of 1a,b with propargyl alcohol 2a and homopropargyl alcohol 2b was carried out in the presence of diisopropyl azodicarboxylate (DIAD) and triphenylphosphine and afforded acetylenic thioimidates 3a–d in 78–92% yield (Scheme 1).

![Scheme 1](image)

**Scheme 1.** Synthesis of acetylenic thioimidates 3a–d and their “click” reaction with AZT (4). (MSA = methanesulfonic acid, TfOH = trifluoromethanesulfonic acid, DIAD = diisopropyl azodicarboxylate; Fc = ferrocenyl; Pyr = 1-pyrenyl).
The $^{13}$C NMR spectra of 3a–d displaying characteristic signals at $\delta$ 171–175 and $\delta$ ~161 confirm the S-propargylation and homopropargylation. The IR spectra of 3a–d show strong absorption bands at 1700–1725 and 1600–1620 cm$^{-1}$, indicative of N-ethoxycarbonylthioimidates.

To test the “clickability” of 3a–d, we chose 3′-azido-3′-deoxythymidine (AZT, 4) as a model azide-modified biomolecule. Modification of nucleosides using the Cu(I)-catalyzed Huisgen-Meldal azide-alkyne cycloaddition (CuAAC) reaction has attracted considerable interest; numerous compounds obtained in this way exhibit interesting biological or pharmaceutical properties. Furthermore, the 1,2,3-triazole formed in CuAAC reactions is now considered a promising pharmacophore. AZT (4) is a commercially available and cheap azide-containing nucleoside, which has been used in “click” reactions with various terminal acetylenes affording conjugates displaying interesting biological properties. However, to the best of our knowledge, up to now there is no report on the reaction of 4 with ferrocenyl or pyrenyl acetylenes.

We have found that AZT (4) reacted with 3a–d under the usual CuAAC reaction conditions to afford 5a–d in 40–70% yield. The formation of the 1,4-disubstituted 1,2,3-triazole ring was confirmed by the $^1$H NMR signals of the H-5 triazolyl protons at $\delta$ 7.5–8.3, and by the absence of acetylenic proton signals at $\delta$ 2–2.5.

### Redox properties of ferrocenyl-AZT conjugates 5a–b

Since ferrocenyl groups are widely used as redox-active reporters in biochemistry, it seemed interesting to study the redox properties of 5a,b. Cyclic voltammetry showed that the Fe(II)/Fe(III) redox process in these compounds is electrochemically quasi-reversible ($\Delta E \sim$80–100 mV) and chemically reversible ($i_{pa}/i_{pc} \sim 1$) occurring at substantially higher potential than that of the parent ferroce (Table 1).

<table>
<thead>
<tr>
<th>Compound</th>
<th>$E_{1/2}$ (mV)$^a$</th>
<th>$\Delta E$ (mV)</th>
</tr>
</thead>
<tbody>
<tr>
<td>5a</td>
<td>282</td>
<td>80</td>
</tr>
<tr>
<td>5b</td>
<td>265</td>
<td>84</td>
</tr>
<tr>
<td>FcAc</td>
<td>248</td>
<td>102</td>
</tr>
</tbody>
</table>

Compounds 5a,b undergo oxidation at slightly higher potential than acetylferrocene, indicating that the ($N$-ethoxycarbonyl)thioimidato groups in these compounds behave as relatively strong electron-withdrawing groups (stronger than the acetyl group). This may be explained by delocalization of the negative charge on the carbonyl group and the cation stabilizing ability of the ferrocenyl group (Scheme 2).
Scheme 2. Resonance structures of a ferrocenyl \((N\text{-ethoxycarbonyl})\text{thioimidate (Z-isomer shown).}\)

**Electronic absorption and emission spectra of 5c,d**

Electronic absorption and emission spectra of 5c and 5d are closely similar. Those of 5c are shown in Fig. 1 and the data are collected in Table 2.

(a)

(b)

*Figure 1.* (a) Electronic absorption and (b) fluorescence emission spectra \((\lambda_{\text{excit}} = 345 \text{ nm})\) of 5c in various solvents.

The electronic absorption spectra of 5c,d exhibit typical vibronic features of the pyrenyl chromophore\(^3\) attributable to the \(S_0\text{–}S_1\) transitions.

The fluorescence displayed by these compounds is strongly solvent-dependent (Table 2). In a nonpolar solvent like benzene, a relatively weak, structured fluorescence spectrum is observed, whereas in other solvents stronger and red-shifted unstructured signals were also present. These signals were observed even at concentrations as low as \(10^{-7}\) M, suggesting that they correspond to electronic transitions in monomers 5c,d. The strongest fluorescence was observed in chloroform and in dichloromethane (DCM). For 5c, the fluorescence quantum yield of 0.04 was determined (in DCM, excitation wavelength 376 nm, quinine sulfate in 0.1 M H\(_2\)SO\(_4\) used as a reference\(^1\)). The solvent-dependence of fluorescent properties of 5c,d suggests that pyrenyl
thioimidato compounds may be used, similarly to pyrenyl carbonyl compounds,\textsuperscript{12} as environment-sensitive probes in biological systems and in materials science.

Table 2. Electronic absorption spectra of 5c,d in various solvents

<table>
<thead>
<tr>
<th>5</th>
<th>Solvent</th>
<th>Absorption $\lambda_{max}$ [nm] ($\varepsilon_{max}$ [M$^{-1}$cm$^{-1}$])</th>
<th>Fluorescence $\lambda_{max}$ [nm]</th>
</tr>
</thead>
<tbody>
<tr>
<td>5c</td>
<td>Benzene</td>
<td>377 (890), 348 (19280), 332 (12790), 316 (6600)</td>
<td>387, 408, 432</td>
</tr>
<tr>
<td></td>
<td>Chloroform</td>
<td>376 (3200), 348 (33390), 332 (22370), 317 (11070)</td>
<td>501</td>
</tr>
<tr>
<td></td>
<td>Dichloromethane</td>
<td>376 (2890), 346 (40590), 331 (27610), 316 (13490)</td>
<td>416, 496</td>
</tr>
<tr>
<td></td>
<td>DMF</td>
<td>376 (1470), 347 (28090), 331 (18610), 317 (7610)</td>
<td>386, 411, 489</td>
</tr>
<tr>
<td></td>
<td>Acetonitrile</td>
<td>375 (1940), 344 (37690), 329 (25150), 314 (12650)</td>
<td>386, 410, 494</td>
</tr>
<tr>
<td></td>
<td>Methanol</td>
<td>375 (2030), 344 (33700), 329 (22200), 316 (9630)</td>
<td>385, 542</td>
</tr>
<tr>
<td>5d</td>
<td>Benzene</td>
<td>376 (1660), 347 (24150), 332 (16370), 316 (7300)</td>
<td>386, 450</td>
</tr>
<tr>
<td></td>
<td>Chloroform</td>
<td>376 (3010), 347 (35030), 331 (23440), 318 (10370)</td>
<td>498</td>
</tr>
<tr>
<td></td>
<td>Dichloromethane</td>
<td>376 (2380), 346 (33760), 331 (22730), 317(10000)</td>
<td>414, 435, 496</td>
</tr>
<tr>
<td></td>
<td>DMF</td>
<td>376 (2310), 346 (29930), 331 (20150), 317(9160)</td>
<td>386, 411, 487</td>
</tr>
<tr>
<td></td>
<td>Acetonitrile</td>
<td>375 (2500), 344 (39160), 329 (26270), 314 (13130)</td>
<td>387, 412, 494</td>
</tr>
<tr>
<td></td>
<td>Methanol</td>
<td>375 (2460), 344 (36400), 329 (24380), 316 (11340)</td>
<td>384, 528</td>
</tr>
</tbody>
</table>

Ferrocenyl compounds 5a,b are not fluorescent (it is known that ferrocene is an efficient quencher of excited states).\textsuperscript{13}

Conclusions

Ferrocenylthioamide 1a is an efficient labeling reagent for hydroxyl-containing biomolecules via Mitsunobu reaction; it can be transformed into ferrocenylthioimidato acetylenes 3a,b that are “clickable” with biomolecules bearing an azido function (exemplified by AZT, 4) affording redox-active 5a,b. The same approach applied to 1-pyrenylthioamide 1b opens an entry to bioconjugates 5c,d with a new environmentally sensitive pyrenylthioimidato fluorophore.

Experimental Section

General. Compound 1a was prepared according to a published procedure.\textsuperscript{6} All other reagents were purchased from Sigma-Aldrich and were used as such. Before use, solvents were thoroughly purified using standard methods. Reactions with ferrocenyl compounds were carried out under argon. Chromatographic separations were performed on silica gel Merck 60 (230–400 mesh ASTM). NMR spectra were recorded in CDCl$_3$ or DMSO-$d_6$ on a Bruker Avance III 600
spectrometer (600 MHz for $^1$H) and referenced to internal TMS. IR spectra were recorded on a FT-IR NEXUS (Thermo Nicolet) spectrometer. Electronic absorption and fluorescence spectra were run on a Perkin Elmer LS 55 spectrofluorimeter. Cyclic voltammetry experiments were carried out on an AUTOLAB (Eco Chimie BV) apparatus in a three electrode system, the working electrode was a Pt disk (1.5 mm diameter), the reference electrode was a ferrocene electrode, and the counter electrode was a cylindrical Pt gauze. These experiments were carried out using argon-saturated 5×10^{-4} M solutions of 5a,b in acetonitrile containing 0.1 M [(n-Bu)_4N][PF_6] as supporting electrolyte. Elemental analyses were performed by Analytical Services of the Center of Molecular and Macromolecular Studies of the Polish Academy of the Sciences (Łódź).

**Ethyl pyrene-1-carbonothioylcarbamate (1b).** To a solution of pyrene (202 mg, 1 mmol) in DCM (5 mL) were added at rt ethoxycarbonylisothiocyanate (262 mg, 2 mmol) and TfOH (348 μL, 4 mmol). After stirring for 2 h, the reaction mixture was poured onto ice-water (30 mL) and extracted with DCM. Flash chromatography (DCM) afforded yellow crystals 1b (316 mg, 95%); mp 159–160 °C. $^1$H NMR (600 MHz, CDCl$_3$): δ 9.48 (s, 1H, NH), 8.24 (d, J = 9 Hz, 1H), 8.21 (dd, J$_1$ = 3.6 Hz, J$_2$ = 11.4 Hz, 2H), 8.16 (d, J = 7.8 Hz, 1H), 8.17 (dd, J$_1$ = 1.8 Hz, J$_2$ = 10.8 Hz, 2H), 8.05 (d, J = 9 Hz, 1H), 8.03 (t, J = 7.8 Hz, 1H), 8.01 (d, J = 7.8 Hz, 1H), 4.04 (q, J = 6.6 Hz, 2H), 1.08 (t, J = 7.2 Hz, 3H). $^{13}$C NMR (150 MHz, CDCl$_3$): δ 201.5, 149.5, 137.2, 132.3, 131.4, 130.7, 128.8, 128.5, 127.3, 126.4, 126.3, 126.0, 125.7, 124.8, 124.6, 124.3, 123.3, 62.9, 13.9. IR (KBr): 3137, 2961, 1762, 1509, 1247, 1235, 1168, 1043, 847 cm$^{-1}$. Anal. calcd for C$_{20}$H$_{15}$NO$_2$: C, 72.05; H, 4.53; N, 4.13. Found: C, 72.01; H, 4.63; N, 4.37%.

**Synthesis of 3a–d. General procedure**

To a stirred solution of 1a or 1b (1.0 mmol) in anhydrous THF (10 mL) under argon at 0 °C were added triphenylphosphine (393 mg, 1.5 mmol), propargyl alcohol 2a or homopropargyl alcohol 2b (1.0 mmol) in THF (5 mL), and DIAD (295 μL, 1.5 mmol). The reaction mixture was allowed to warm to rt and was stirred for 2 h. Flash chromatography (DCM) afforded products 3.

**Prop-2-yn-1-yl N-ethoxycarbonylpyrene-1-carbimidothioate (3a).** Red oil (277 mg, 78%). $^1$H NMR (600 MHz, CDCl$_3$): δ 4.65 (t, J = 1.8 Hz, 2H), 4.31 (t, J = 1.8 Hz, 2H), 4.27 (s, 5H), 4.23 (q, J = 6.6 Hz, 2H), 3.79 (d, J = 2.4 Hz, 2H), 2.23 (t, J = 2.4 Hz, 1H), 1.30 (t, J = 7.2 Hz, 3H). $^{13}$C NMR (150 MHz, CDCl$_3$): δ 171.1, 161.4, 78.5, 71.1, 71.4, 71.2, 70.9, 68.9, 62.5, 19.6, 14.3. IR (KBr): 3287, 2964, 2926, 1699, 1618, 1254 cm$^{-1}$. Anal. calcd for C$_{17}$H$_{17}$FeNO$_2$: C, 57.48; H, 4.82; N, 3.94. Found: C, 57.58; H, 4.91; N, 3.92.

**But-3-yn-1-yl N-ethoxycarbonylpyrene-1-carbimidothioate (3b).** Red oil (321 mg, 87%). $^1$H NMR (600 MHz, CDCl$_3$): δ 4.65 (t, J = 1.8 Hz, 2H), 4.13 (t, J = 1.8 Hz, 2H), 4.25 (s, 5H), 4.23 (q, J = 6.6 Hz, 2H), 3.16 (t, J = 7.2 Hz, 2H), 2.61 (td, J$_1$ = 2.4 Hz, J$_2$ = 7.8 Hz, 2H), 2.04 (t, J = 2.4 Hz, 1H), 1.30 (t, J = 7.2 Hz, 3H). $^{13}$C NMR (150 MHz, CDCl$_3$): δ 171.8, 161.6, 82.4, 77.7, 71.0, 70.9, 69.6, 69.0, 62.4, 29.9, 18.8, 14.3. IR (KBr): 3292, 3097, 2980, 2936, 1705, 1616, 1253 cm$^{-1}$. Anal. calcd for C$_{18}$H$_{19}$FeNO$_2$: C, 58.55; H, 5.19; N, 3.79. Found: C, 58.72; H, 5.22; N, 3.89.
Prop-2-yn-1-yl N-ethoxycarbonylferrocenecarbimidothioate (3c). Colorless crystals (342 mg, 92%); mp 105–106 °C. 1H NMR (600 MHz, CDCl3): δ 8.52 (d, J 9 Hz, 1H), 8.22 (dd, J1 4.8 Hz, J2 10.8 Hz, 2H), 8.16 (d, J 9 Hz, 1H), 8.15 (d, J 7.8 Hz, 1H), 8.11 (d, J 9 Hz, 1H), 8.04 (d, J 9 Hz, 1H), 8.03 (d, J 7.8 Hz, 1H), 7.99 (d, J 7.8 Hz, 1H), 4.05 (d, J 2.4 Hz, 2H), 3.80 (q, J 7.2 Hz, 2H), 2.33 (t, J 2.4 Hz, 1H), 0.73 (t, J 7.2 Hz, 3H). 13C NMR (150 MHz, CDCl3): δ 174.8, 160.6, 132.6, 131.1, 130.7, 129.5, 128.8, 128.7, 127.6, 127.0, 126.4, 126.0, 125.9, 125.0, 124.4, 124.2, 124.1, 124.0, 77.9, 71.9, 62.4, 20.5, 13.7. IR (KBr): 3277, 2981, 1720, 1617, 1220, 846 cm⁻¹. Anal. calc'd for C23H17NO2S: C, 74.37; H, 4.61; N, 3.77. Found: C, 74.45; H, 4.71; N, 3.87.

But-3-yn-1-yl N-ethoxycarbonylferrocenecarbimidothioate (3d). Light yellow crystals (343 mg, 89%); mp 92–94 °C. 1H NMR (600 MHz, CDCl3): δ 8.23 (d, J 9 Hz, 3H), 8.15 (dd, J1 4.2 Hz, J2 12.6 Hz, 3H), 8.06 (d, J 9 Hz, 1H), 8.05 (t, J 7.8 Hz, 1H), 7.97 (d, J 7.8 Hz, 1H), 3.76 (q, J 7.2 Hz, 2H), 2.42 (t, J 7.2 Hz, 2H), 2.61 (td, J1 2.4 Hz, J2 7.2 Hz, 2H), 2.11 (t, J 2.4 Hz, 1H), 0.70 (t, J 7.2 Hz, 3H). 13C NMR (150 MHz, CDCl3): δ 175.9, 160.9, 132.5, 131.1, 130.8, 130.5, 128.8, 128.6, 127.4, 127.1, 126.4, 125.9, 125.8, 125.0, 124.4, 124.3, 124.1, 82.0, 70.0, 62.3, 30.8, 18.6, 13.7. IR (KBr): 3291, 3048, 2977, 1723, 1610, 1236, 846 cm⁻¹. Anal. calc'd for C24H19NO2S: C, 74.78; H, 4.97; N, 3.63. Found: C, 74.82; H, 4.90; N, 3.67%.

Synthesis of 5a–d via “click” reaction. General procedure
To a solution of 3a–d (0.29 mmol) and AZT (4) (90 mg, 0.34 mmol, 1.16 equiv.) in methanol/water (20:2 mL) were added aqueous solutions of TTTA (0.01 M, 250 µL), CuSO4 (0.1 M, 50 µL) and sodium ascorbate (0.1 M, 50 µL). The resulting solution was stirred at rt for 72 h. Then, water (50 mL) was added and products were extracted with ethyl acetate. Pure products were obtained by chromatography using DCM / ethyl acetate / methanol 25:25:1 (v/v/v) as eluent.

5a. Orange crystals (80 mg, 42%); mp 130–131 °C. 1H NMR (600 MHz, DMSO-d6): δ 11.34 (br. s., 1H), 8.22 (s, 1H), 7.82 (br. s., 1H), 6.42 (t, J 6.4 Hz, 1H), 5.37 (d, J 7.5 Hz, 1H), 5.28 (br s, 1H), 4.63 (br. s., 2H), 4.58 (br s, 2H), 4.29 (s, 2H), 4.25 (s, 5H), 4.23–4.16 (m, 3H), 3.76–3.67 (m, 1H), 3.63 (d, J 11.7 Hz, 1H), 2.73 (d, J 6.4 Hz, 1H), 2.69–2.60 (m, 1H), 1.82 (s, 3H), 1.26 (t, J 7.0 Hz, 3H). 13C NMR (150 MHz, DMSO-d6): δ 170.78, 164.17, 161.10, 150.90, 143.08, 136.68, 123.77, 110.08, 84.93, 84.41, 71.98, 71.15, 69.10, 62.53, 61.18, 59.68, 37.59, 25.77, 21.22, 14.62, 12.69, IR IR (KBr): 3432, 1697, 1236 cm⁻¹. Anal. calc'd for C27H30FeN6O6S CH2OH: C, 51.38; H, 5.24; N, 12.84. Found: C, 51.58; H, 5.54; N, 13.12%.

5b. Orange crystals (76 mg, 41%); mp 163–166 °C. 1H NMR (600 MHz, CDCl3): δ 9.20 (br. s., 1H), 7.58 (s, 1H), 7.50 (s, 1H), 6.24 (t, J 6.2 Hz, 1H), 5.42–5.33 (m, 1H), 4.60 (br s, 2H), 4.41 (br s, 3H), 4.29–4.18 (m, 7H), 3.96 (d, J 11.7 Hz, 1H), 3.79–3.65 (m, 2H), 3.36 (qd, J1 6.9 Hz, J2 17.4 Hz, 2H), 3.14 (t, J 7.0 Hz, 2H), 3.01–2.84 (m, 2H), 1.32 (t, J 7.2 Hz, 3H), 1.26 (br s, 3H). 13C NMR (150 MHz, CDCl3): δ 172.63, 163.77, 161.96, 161.96, 150.42, 146.20, 137.65, 121.67, 111.13, 88.21, 85.22, 77.69, 71.18, 70.93, 68.95, 62.57, 61.41, 58.97, 37.56, 29.96, 25.44, 14.33, 12.41. IR IR (KBr): 3437, 1697, 1670, 1236 cm⁻¹. Anal. calc'd for C28H32FeN6O6S: C, 52.10; H, 5.15; N, 13.02. Found: C, 52.47; H, 5.19; N, 13.11%.
5c. Pale yellow crystals (80 mg, 41%); mp 147–149 °C. $^1$H NMR (600 MHz, CDCl$_3$): 9.52 (s, 1H), 8.08 (dd, $J_1$ 4.3 Hz, $J_2$ 7.3 Hz, 2H), 8.02–7.94 (m, 4H), 7.92–7.87 (m, 2H), 7.84 (s, 1H), 7.80 (d, $J$ 7.9 Hz, 1H), 7.41 (s, 1H), 6.19 (t, $J$ 6.2 Hz, 1H), 5.44–5.33 (m, 2H), 4.46 (d, $J$ 2.6 Hz, 2H), 4.35 (br s, 2H), 3.93 (d, $J$ 11.3 Hz, 1H), 3.76 (d, $J$ 11.3 Hz, 1H), 3.67 (q, $J$ 6.9 Hz, 2H), 2.96–2.87 (m, 2H), 2.84 (d, $J$ 6.0 Hz, 1H), 1.76 (s, 3H), 0.57 (t, $J$ 7.0 Hz, 3H).

$^{13}$C NMR (150 MHz, CDCl$_3$): δ 175.6, 164.0, 160.9, 150.5, 143.7, 137.6, 132.5, 131.0, 130.5, 129.6, 128.8, 128.6, 127.3, 126.9, 126.4, 126.0, 125.9, 124.9, 124.2, 124.1, 124.0, 123.9, 123.1, 111.0, 88.0, 85.2, 62.5, 61.4, 59.3, 37.4, 26.7, 13.6, 12.3. IR (KBr): 3432, 1705, 1236 cm$^{-1}$.

Anal. calcd for C$_{33}$H$_{30}$N$_6$O$_6$S$\cdot$CH$_3$OH: C, 60.88; H, 5.11; N, 12.53. Found: C, 60.43; H, 5.73; N, 12.93%.

5d. Pale yellow crystals (152 mg, 71%); mp 182–183 °C. $^1$H NMR (600 MHz, DMSO-d$_6$): $\delta$ 11.27 (s, 1H), 8.34–8.30 (m, 2H), 8.27 (d, $J$ 9.4 Hz, 1H), 8.27 (d, $J$ 7.9 Hz, 1H), 7.89 (d, $J$ 7.9 Hz, 1H), 7.75 (s, 1H), 6.38 (t, $J$ 6.8 Hz, 1H), 5.30 (td, $J$ 5.4, 8.5 Hz, 1H), 5.19 (br. s., 1H), 4.19–4.14 (m, 1H), 3.68–3.60 (m, 2H), 3.59–3.53 (m, 1H), 3.47 (t, $J$ 6.8 Hz, 2H), 3.24 (br. s., 1H), 3.15 (t, $J$ 7.2 Hz, 2H), 2.75–2.65 (m, 1H), 2.63–2.50 (m, 1H), 1.78 (s, 3H), 0.49 (t, $J$ 7.2 Hz, 3H). $^{13}$C NMR (150 MHz, DMSO-d$_6$): δ 175.7, 164.2, 160.6, 150.9, 145.5, 136.7, 132.4, 131.1, 130.7, 130.6, 129.3, 129.1, 127.6, 127.4, 127.1, 126.8, 126.6, 125.4, 124.9, 124.3, 123.9, 123.8, 122.7, 110.1, 85.0, 84.4, 62.2, 61.3, 59.6, 37.6, 31.3, 25.1, 14.0, 12.7. IR (KBr): 3432, 1705, 1236 cm$^{-1}$. Anal. calcd for C$_{34}$H$_{32}$N$_6$O$_6$S$\cdot$CH$_3$COOC$_2$H$_5$: C, 62.97; H, 5.56; N, 11.59 Found: C, 63.12 ; H, 5.50; N, 11.98%.

Acknowledgements

Financial support from the Polish Committee for Scientific Research (KBN) (research project N N204 154636) is gratefully acknowledged.

References


