Incorporating Thiobarbituric Acid with 1,2,4-Triazin-6-one Containing Nitroarginine Moiety

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Abstract

Nitrilimines (2a,b), react with nitroarginine methyl ester (3) at room temperature, through cyclocondensation reaction, to give 1-aryl-3-substituted-1,2,4-triazin-6-ones (4a,b). Condensation of these compounds with thiosemicarbazide give the thiosemicarbazone derivatives (5a,b) which upon heterocyclization with malonic acid give the thiobarbituric acid derivatives (6a,b). The structures of these compounds were deduced from: IR, mass, \textsuperscript{1}H and \textsuperscript{13}C NMR spectra.

Keywords Nitrilimine, nitroarginine methyl ester, 1,2,4-triazin-6-ones, thiosemicarbazone, thiobarbituric acid derivatives.

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1. Introduction

1,2,4-Triazines play a vital role in many biological processes and synthetic drugs. Furthermore, many heterocyclic systems bearing 1,2,4-triazines exhibit remarkable pharmacological effects [1-5]. In search for new anti-HIV and anticancer agents, some additional heterocyclic moieties were incorporated in the 1,2,4-triazine nucleus via the interaction between functionalized 1,2,4-triazines with various nucleophilic and electrophilic reagents in different media [6]. A lot of information about the important methods of synthesis, structure, physical properties and chemical reactivity of these compounds are described in the literature [7-9].

A facile preparation of 1,2,4-triazin-6-ones from the reaction of nitrilimines with α-amino esters was reported [10], as well as the reaction of nitrilimines with α-aminoacetonitrile [11]. Nitrilimines react with α-hydrazinoester to give 1,2,4-triazin-6-ones [12], also the synthesis of 1,2,4-triazin-6-one containing nitroarginine have been reported [13]. Barbituric acid or thiobarbituric acid are well known for their biological activity [14-16]. The incorporation of compounds (4) (scheme 1) and thiobarbituric acid into a single molecule may enhance the activity of the target molecule. The new triazinone derivatives will be bioassayed, and the results will be communicated separately.

Scheme 1  Synthesis of compounds 6
2. Results And Discussion

Nitrilimines (2), generated in situ from the respective hydrazonyl halides (1) in THF upon the addition of triethylamine, are found to react with nitroarginine methyl ester (3) in methanol as a solvent at room temperature, through cyclocondensation reaction, to give 1,2,4-triazin-6-ones (4). Compounds (4) are achieved via a nucleophilic attack of the amino group of the nitroarginine (3) at nitrilimines followed by intracyclization. Synthesis of some new heterobicyclic systems bearing a 1,2,4-triazinone moiety was performed starting from 1,2,4-triazinones (4). Condensation of these compounds with thiosemicarbazide in boiling ethanol and few drops of acetic acid produced the semicarbazon derivative (5). Refluxing of 5 with malonic acid caused competitive heterocyclization on the thiosemicarbazone moiety and yielded the thiobarbituric acid derivative (6).

Structures of compounds (5) and compounds (6) have been deduced from IR, mass, 1H-NMR and 13C-NMR spectra. The IR spectra of compounds (5) in KBr revealed strong bands for all types of (NH) stretching in the region 3450-3280 cm⁻¹. One absorption band is also observed about 1660 cm⁻¹ (C=O triazinone). Three stretching bands of C=N appeared in the region 1640-1610 cm⁻¹. The IR spectra of compounds (6) in KBr revealed two new strong bands around 1670 cm⁻¹ for (C=O) indicating amides produced from reaction of compounds (5) with malonic acid. The structure of compound (6b) for example was also deduced from mass spectrum fragmentation pattern of this compound which shows the molecular ion peak M⁺ 502, the base peak m/e 57 [CH₂CONH]⁺ path a. An important fragmentation is observed (m/e 316) M⁺–186 [COCH(CH₃)₂NHC=NNHNHNO₂]⁺ (path b). Another important fragmentation is observed (m/e 399) M⁺ – 103 [NHC=NNHNHNO₂]⁺ (path c) (Scheme 2).

The 1H-NMR spectra of compounds (5) show a singlet at 10.0 ppm for NH proton of thiosemicarbazon and a singlet at 10.6 ppm for NH₂ protons. The 1H-NMR spectra of compounds (6) show a singlet at 7.3 ppm for NH proton of triazinone and three singlets for 3NH of nitroguanidine are observed between 8.5-7.6 ppm. The NH and CH₂ protons of pyrimidine appear as singlets at 10.4 and 3.15 ppm respectively. 

13C-NMR spectra of compounds (6) show two new signals for CH₂ and C=O of pyrimidine at 45 and 170 ppm, respectively. Compounds (7) contain primary NH₂ and secondary NH groups. However, compounds (6) contain only secondary NH groups. The 1H-NMR shows clearly the triazine ring NH which appears as a singlet at 7.3 ppm, three NHs for nitroarginine as three singlets between 8.5-7.9 ppm and the NH of thiobarbituric acid appear as singlet around 10.4 ppm. The singlet signal of NH of semicarbazon at 9.89 in compound (5) disappeared in compound (6) and the two H’s of NH₂ converted to one hydrogen for NH means that the cyclization occurred on the thiosemicarbazon moiety. The mass spectrum fragmentation pattern of compounds (6) support the the cyclization of malonic acid on the thiosemicarbazon moiety, through the fragmentation pattern b (Scheme 2). These results agree with our expectation of obtaining compound (6) and not (7) due to the deactivating group NO₂ on the guanidine moiety which diminishes its nucleophilicity and consequently prevents the cyclization at that position.
INCORPORATING THIOBARBITURIC ACID.

Scheme 2 Mass fragmentation pattern of compound 6b

3. Experimental

Melting points (uncorrected) were determined on Steuart melting point apparatus. IR spectra (in cm⁻¹) records as KBr discs on Perkin-Elmer 237 infrared spectrometer. Mass spectra were obtained using GCMS-QP10000 EX Schimadzu spectrometre at 70 eV. ¹H and ¹³C-NMR were recorded on a Bruker AM 300 MHz NMR spectrometer using DMSO-d₆ as a solvent at 21°C and TMS as an internal reference. Chemical shifts are expressed in δ (ppm) downfield from TMS and coupling constants are in Hertz (Hz).

The appropriate hydrazonoyl chlorides employed in this work were prepared following standard procedure [17]. Nitroarginine methyl ester hydrochloride (3) employed in this work was obtained by reaction of nitroarginine with thionyl chloride in methanol following standard procedure [13].

1,2,4-triazin-6-ones (4a,b) were obtained by following the standard procedure [13].

Synthesis of (5a, b) :

To a stirred solution of the particular 1,2,4-triazin-6-one (4) (10 mmol) in absolute ethanol (60 mL) acidified with drops of glacial acetic acid was added the thiosemicarbazide and the stirring reaction mixture was refluxed for 2-3 hours. The solvent was then evaporated in vacuo and the residue was collected and recrystallized from ethanol to yield the thiosemicarbazone derivatives (5a,b).

The following compounds were synthesized using this method:
1-(4-Chlorophenyl)-3-(thiosemicarbazonoacetyl)-4,5-dihydro-5-[3 nitroguanidinoprop-1-yl]-1H-1,2,4-triazin-6-one (5a)

Yield = 75%, M. P. = 175-176 °C, IR (KBr): cm⁻¹ 3475-3290 (all NH and NH₂ bands), 1660 (C=O lactam), 1638 - 1590 (3 C=N). ¹H-NMR (DMSO-d₆): δ / ppm 1.56 (m, 2H, CH₂), 1.85 (m, 2H, CH₂), 2.10 (s, 3H, CH₃C=N), 3.13 (m, 2H, CH₂NH), 4.06 (m, 1H, H-5), 7.5 (s, 1H, NH), 7.5-7.4 (2d, 4H aromatic protons, J = 7 Hz), 8.5-7.8 (s, 3H, 3NH nitroguanidine), 9.89 (s, 1H, NH thiosemicarbazone), 10.4 (s, 2H, NH₂). ¹³C NMR (DMSO-d₆): δ / ppm 11.8 (CH₃C=N), 24.3, 30.6 (CH₂), 40.8 (CH₂NH), 53.1 (C=5), 126.3, 128.8, 130.5, 140.3, 143.2 (aromatic carbons ), 145.05 (C-3), 152 (CH₃C=N), 160 (C=N nitroguanidine), 162.7 (C=O lactam), 179.6 (C=S). MS: m/z (C₁₆H₁₃ClN₂O₂S) = 468/470 (M⁺), 267/269 [CH₃C(NNNCSNH₂)C=NN-p-ClC₆H₄]⁺, 113/113 [p-ClC₆H₄N⁺], 103 [NO₂NHCNHNH₂⁺], 90 [N₃H₂CSNHNH₂⁺], 75 [NH₂CSNH₂⁺], 60 [NH₂CS⁺].

1-Phenyl-3-(thiosemicarbazonoacetyl)-4,5-dihydro-5-[3 nitroguanidinoprop-1-yl]-1H-1,2,4-triazin-6-one (5b)

Yield = 77%, M. P. = 179-180 °C, IR (KBr): cm⁻¹ 3475-3280 (all NH bands), 1662 (C=O lactam), 1640 - 1590 (3 C=N). ¹H-NMR (DMSO-d₆): δ / ppm 1.86 (m, 2H, CH₂), 2.00 (m, 2H, CH₂), 2.10 (s, 3H, CH₃C=N), 3.20 (m, 2H, CH₂NH), 4.05 (m, 1H, H-5), 6.85 (s, 1H, NH) 7.40-7.20 (m, 5H aromatic protons), 8.5-7.9 (s, 3H, 3NH nitroguanidine), 10.1 (s, 1H, NH thiosemicarbazone), 10.5 (s, 2H, NH₂). ¹³C NMR (DMSO-d₆): δ / ppm 14.0 (CH₃C=N), 24.3, 30.6, (CH₂), 40.8 (CH₂NH), 53.3 (C=5), 114.2, 123.2, 129.9, 138.4 (aromatic carbons ), 144.0 (C-3), 151.2 (CH₃C=N), 159.7 (C=N nitroguanidine), 162.7 (C=O lactam), 179.4 (C=S). MS: m/z (C₁₆H₁₃N₂O₂S) = 356/358 (M⁺), 233 [CH₃C(NNNCSNH₂)C=NNC₆H₄]⁺, 103 [NO₂NHCNHNH₂⁺], 91 [C₆H₇⁺], 90 [NH₂CSNH₂⁺], 77 [C₆H₅⁺], 57 [CH₃CONH⁺].

Synthesis of (6a,b) :

To a stirred solution of the thiosemicarbazone derivatives (2 mmol) in methanol (30 mL) was added a solution of malonic acid (2.5 mmol) in methanol (30 mL). The resulting reaction mixture was heated at refluxed 4 hours and the solvent was then removed in vacuo. The residue was collected and recrystallized from ethanol to yield (6a,b).

The following compounds were synthesized using this method:

1-(4-arylimino-3-(2’-thio-dihydropyrimidine-4’,6’-dione)ethyldien-4,5-dihydro-5-[3 nitroguanidinoprop-1-yl]-1H-1,2,4-triazin-6-one (6a)

Yield = 76%, M. P. = 200-201 °C, IR (KBr): cm⁻¹ 3480-3250 (3NH nitroguanidine, 1NH triazinone and 1NH pyrimidine), 1665, 1640 (two C=O ), 1635 - 1580 (3 C=N). ¹H-NMR (DMSO-d₆): δ / ppm 1.20 (m, 2H, CH₂), 1.65 (m, 2H, CH₂), 2.00 (s, 3H, CH₃C=N), 3.00 (m, 2H, CH₂NH), 3.15 (s, 2H, CH₂, pyrimidine), 4.06 (m, 1H, H-5), 7.3 (s, 1H, NH triazinone), 7.58 - 7.40 (2d, 4H aromatic protons, J = 7 Hz), 8.5-7.9 (s, 3H, 3NH nitroguanidine), 10.40 (s, 1NH, pyrimidine). ¹³C NMR (DMSO-d₆): δ / ppm 11.7 (CH₃C=N), 46.0 ( CH₂ pyrimidine), 18.1, 30.1, (CH₂), 40.2 (CH₂NH), 53.0 (C=5), 126.3, 128.7, 130.5, 140.0 (aromatic carbons ), 145.3 (C-3), 150.0 (CH₃C=N), 159 (C=N nitroguanidine), 163.0 (C=O triazinone), 170.0 (C=O pyrimidine), 179.6 (C=S). MS: m/z (C₁₉H₁₉ClN₁₀O₅S) = 536/538 (M⁺), (433/435 [C₈H₁₃ClN₁₀O₅S]⁺). M⁺ – 103, (350/352 [C₆H₁₃ClN₁₀O₅S]⁺). M⁺ – 186, 103 [NO₂NHCNHNH₂⁺], 91 [C₆H₇⁺], 77 [C₆H₅⁺], 57 [CH₃CONH⁺].

1-Phenyl-3-(2’-thio-dihydropyrimidine-4’,6’-dione)ethyldien-4,5-dihydro-5-[3 nitroguanidinoprop-1-yl]-1H-1,2,4-triazin-6-one (6b)

Yield = 84%, M. P. = 202-204 °C, IR (KBr): cm⁻¹ 3480-3250 (3NH nitroguanidine, 1NH triazinone and 1NH pyrimidine), 1665 (C=O amide), 1655 (C=O lactam), 1640 - 1590 (3 C=N). ¹H-NMR (DMSO-d₆): δ / ppm 1.55 (m, 2H, CH₂), 1.70 (m, 2H, CH₂), 2.00 (s, 3H,
CH₃(C=N), 3.16 (m, 2H, CH₂NH), 3.20 (s, 2H, CH₂, pyrimidine), 4.10 (m, 1H, H-5), 7.2 (s, 1H, NH triazinone), 7.52 – 6.98 (m, 5H aromatic protons), 8.5-7.8 (s, 3H, 3NH nitroguanidine), 10.50 (s, 1NH, pyrimidine).

13C NMR (DMSO-d₆): δ / ppm 14.1 (CH₃C=N), 24.0, 30.0, (CH₂), 40.0 (CH₂NH), 45.0 (CH₂ pyrimidine), 53.0 (C-5), 126.2, 128.9, 130.5, 140.3 (aromatic carbons ), 143.60 ( C=N), 151.00 (CH₃C=N), 159.00 (C=N nitroguanidine), 162.81 (C=O triazine), 170.00 (C=O pyrimidine), 179.40 (C=S). MS: mz (C₁₅H₁₀N₄O₅S) = 502 (M⁺), (399) [C₁₅H₁₀N₄O₅S]⁺ M⁺–103, (316) [C₁₅H₁₀N₄O₅S]⁺ M⁺–186, 103 [NO₂NHCNHNH²⁺], 91 [C₆H₅N⁺], 77 [C₆H₃⁺], 57 [CH₃CONH]⁺.

References

