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Studies on some N-bridged heterocycles derived from bis-[4-amino-5-mercapto-1,2,4-triazol-3-yl] alkanes

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Abstract

A series of bis-[4-amino-5-mercapto-1,2,4-triazol-3-yl] alkanes have been synthesized and were converted into bis-[1,2,4-triazolo[3,4-b]-1,3,4-thiadiazol-4-yl] alkanes. The newly synthesized compounds were characterized by analytical IR, NMR and mass spectral studies. Some of the newly synthesized compounds were screened for their antibacterial properties and exhibited activity with MIC in the range 3–12.5 μg/ml.

Keywords: N-bridged heterocycles; Bis-[4-amino-5-mercapto-1,2,4-triazol-3-yl] alkanes; Antibacterial properties

1. Introduction

Various 1,2,4-triazoles and the N-bridged heterocycles derived from them are found to be associated with diverse pharmacological activities [1–10]. A number of 1,3,4-thiadiazoles showed antibacterial properties similar to those of well known sulphonamide drugs [11]. Thus, the thiadiazole nucleus which incorporates a N–C–S linkage exhibits a large number of biological activities [12]. Prompted by the biological activities of triazoles and N-bridge heterocycles derived from them [13–18], a number of bis-[4-amino-5-mercapto-1,2,4-triazol-3-yl] alkanes were prepared and cyclized with various carboxylic acids in the presence of phosphorus oxychloride to yield triazolothiadiazolyl alkanes. Some of the selected compounds were screened for their antibacterial activities. Results of such studies are described in this paper.

2. Chemistry

Three different dicarboxylic acids were fused with thiocarbohydrazide to obtain bis-[4-amino-5-mercapto-1,2,4-triazol-3-yl] alkanes (**3**) (Scheme 1). Formation of these bis-triazolyl alkanes was confirmed by elemental analysis and spectral studies [19]. Cyclization of these triazolyl alkanes with substituted aromatic carboxylic acids (**4**), substituted aryloxyacetic acids (**6**) and substituted anilinoacetic acids (**8**) was carried out using phosphorous oxychloride as a cyclizing agent (Scheme 1). Compounds **6** were prepared from substituted phenols and monochloroacetic acid [20]. Compounds **8** were prepared from substituted anilines and monochloroacetic acid [20]. The cyclization of triazolyl alkanes with various carboxylic acids yielded the condensed triazolo thiadiazolyl alkanes (**5**, **7** and **9**). The structures of **5**, **7** and **9** were confirmed on the basis of elemental analysis, IR, NMR and mass spectral data. The characterization data of these condensed heterocycles are given in Tables 1–3.

All the newly synthesized compounds were analyzed satisfactorily for their nitrogen content. The IR spectrum of **5a** showed an absorption band at 1593 and 1490 cm^{-1} indicating the presence of C=N and C=C in the ring. In the IR spectra of the cyclized products the absorption bands at 3100–3200 cm^{-1} attributed to the NH functional group were absent and also the absorption band at 2360 cm^{-1} attributed to the SH functional group was absent. This confirmed the involvement of the NH_2 and SH groups of the parent bis-triazole in the ring formation.

The formation of cyclized products **5**, **7** and **9** were further supported by recording the NMR spectra of a few selected compounds. In the ^1H NMR spectrum of bis-triazolothiadiazolyl alkanes, the characteristic downfield signal at δ 13.4 attributable to the –N=C–SH moiety was absent. A sharp singlet at δ 5.4 attributable to the N–NH_2 group in the parent bis-triazolyl alkanes was also absent in the cyclized product. In the ^1H NMR spectrum of **5c**, a triplet at δ 2.9 was attrib-

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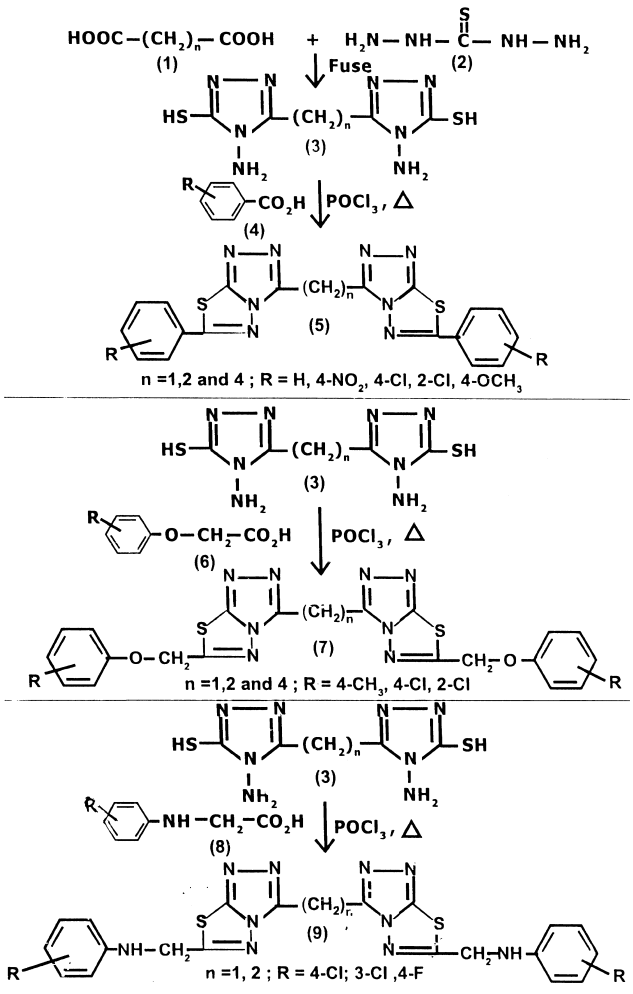


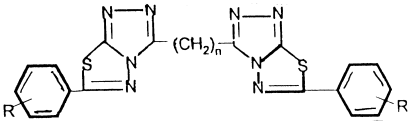
Scheme 1.

uted to the four methylene protons of the alkyl chain while the aromatic protons of the 4-nitro benzene ring appeared as two doublets at δ 8.2 ($J = 10$ Hz) and 8.4 ($J = 10$ Hz). The ^{1}H NMR spectrum of **7c** showed a triplet at δ 1.9 integrating for four protons, characteristic of the four central methylene protons while a similar triplet at δ 3.1 integrating for four protons is attributable to the four remaining methylene protons of the alkyl chain, etc. A sharp singlet at δ 5.5 integrating for four protons is attributed to the $-OCH_2$ groups. The aromatic protons of the chlorophenyl moiety appeared as two doublets at δ 7.2 (8 Hz) and 7.4 (8 Hz). The ^{1}H NMR spectral data of cyclized compounds are given in Tables 1 and 2.

The mass spectra of two cyclised products **5a** and **5e** were recorded and were found to be consistent with the assigned structures. Molecular ions of the triazolo thiadiazolyl alkanes (**5a**) and (**5e**) were observed at $m/z = 458, 526$, etc. corresponding to the molecular formulae $C_{22}H_{18}N_8S_2$ and $C_{22}H_{16}Cl_2N_8S_2$ respectively. M + 2 and M + 4 peaks were observed in **5e**, which are attributed to the presence of two chlorine atoms. The molecular ion of **5a** underwent fragmentation to produce a molecular ion at $m/z = 355$, which corresponds to the loss of benzonitrile. A base peak at $m/z = 229$ indicates the presence of doubly-charged molecular ion (A). The peak at $m/z = 121$, was attributed to the ion (B) which further underwent fragmentation to give phenyl cation at $m/z = 77$. The mass spectral fragmentation of the cyclized product **5a** is given in Scheme 2.

The mass spectra of two cyclized products **7b** and **7f** were also recorded and were found to be consistent with the assigned structures. Molecular ions of the cyclized products **7b** and **7f** were observed at $m/z = 558/560/562$ correspond-

Table 1
Bis-[6-aryl-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazol-4-yl] alkanes (**5a–h**)[a]



Compound no.	R	*n*	% Yield	M.p. (°C)	Nature of the compound	Molecular formula	% N found (calc.)
5a[b]	H	4	74	228–230	pale yellow crystals	$C_{22}H_{18}N_8S_2$	24.65 (24.62)
5b	4-NO_2	1	76	264–266	light yellow needles	$C_{19}H_{10}N_{10}O_4S_2$	27.72 (27.6)
5c[c]	4-NO_2	2	78	176–178	pale yellow micro crystals	$C_{20}H_{12}N_{10}O_4S_2$	27.03 (26.9)
5d[d]	4-NO_2	4	76	174–176	pale yellow crystals	$C_{22}H_{16}N_{10}O_4S_2$	25.48 (25.5)
5e[e]	4-Cl	4	81	298–300	white micro crystals	$C_{22}H_{16}Cl_2N_8S_2$	21.42 (21.3)
5f	2-Cl	2	83	> 300	pale yellow crystals	$C_{20}H_{12}Cl_2N_8S_2$	22.39 (22.5)
5g	2-Cl	4	80	150–152	pale yellow micro crystals	$C_{22}H_{16}Cl_2N_8S_2$	21.28 (21.3)
5h	4-OCH_3	4	68	88–90	pale yellow micro crystals	$C_{24}H_{22}N_8O_2S_2$	21.52 (21.6)

[a] Solvent of crystallization, DMF.

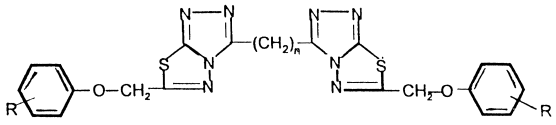
[b] MS: *m/e* (% abundance) 458 (10.5, M^+), 355 (44.1, M-ArCN), 229 (100, A) 121 (72.4, B), 77 (80, $C_6H_5^+$); IR (KBr): 3066 cm^{-1} (γ_{C-H} ArH), 1590 cm^{-1} ($\gamma_{C=N}$), 1313 cm^{-1} (γ_{CN}).

[c] ^{1}H NMR (DMSO-d_6): δ 8.4 (d, 4H, $J = 10$ Hz, aromatic protons), 8.2 (d, 4H, $J = 10$ Hz, aromatic protons), δ 2.9 (br, t, 4H, $-CH_2$).

[d] MS: *m/e* (% abundance) 502 (1.2, M-NO_2), 73 (100).

[e] MS: *m/e* (% abundance) 526 (21.7, M^+), 95 (100); IR (KBr): 3078 cm^{-1} (γ_{C-H} ArH), 1593 cm^{-1} ($\gamma_{C=N}$), 1403 cm^{-1} (γ_{CN}).

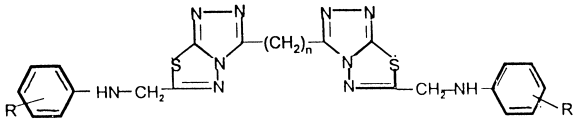
Table 2
Bis-[6-aryloxymethyl-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazol-4-yl] alkanes (**7a–f**)[a]



Compound no.	R	*n*	% Yield	M.p. (°C)	Nature of compound	Molecular formula	% N found (calc.)
7a	4-Cl	1	82	223–225	light yellow needles	$C_{21}H_{14}Cl_2N_8O_2S_2$	20.52 (20.6)
7b[b]	4-Cl	2	78	260–262	white micro crystals	$C_{22}H_{16}Cl_2N_8O_2S_2$	20.14 (20.00)
7c[c]	4-Cl	4	76	235–238	white micro needles	$C_{24}H_{20}Cl_2N_8O_2S_2$	19.04 (19.1)
7d	4-CH_3	2	72	148–150	light yellow micro crystals	$C_{24}H_{22}N_8O_2S_2$	21.47 (21.6)
7e	4-CH_3	4	71	100–102	light yellow needles	$C_{26}H_{26}N_8O_2S_2$	20.62 (20.5)
7f[d]	2-Cl	2	76	218–220	Creemish white crystals	$C_{26}H_{16}C_{12}N_8O_2S_2$	20.17 (20.0)

[a] Solvent of crystallization, DMF.
[b] MS: *m/e* (% abundance) 558 (3.2, M^+), 432 (3.1, M-$ClC_6H_5O^+$), 167 (5.9, C), 128 (36.2), 99 (49.4), 64 (28.1), 46 (100). IR (KBr): 3091 cm^{-1} (γ_{C-H} ArH), 1587 cm^{-1} ($\gamma_{C=N}$), 1283 cm^{-1} (γ_{C-N}). ^{1}H NMR (DMSO-d_6): δ 7.4 (d, 4H, $J = 8$ Hz, aromatic protons), δ 7.2 (d, 4H, $J = 8$ Hz, aromatic protons), δ 2.5 (s, 2H, $-CH_2$).
[c] ^{1}H NMR (DMSO-d_6): δ 7.4 (d, 4H, $J = 8$ Hz, aromatic protons), δ 7.2 (d, 4H, $J = 8$ Hz, aromatic protons), δ 5.5 (s, 4H, $-OCH_2$), δ 3(br, t, 4H, $-CH_2$). δ 1.9 (br, t, 4H, $-CH_2$).
[d] MS: *m/e* (% abundance) 558 (3.4 M^+), 432 (3.1, M-$ClC_6H_5O^+$), 167 (10.9, C), 128 (100), 99 (34.2), 64 (60.2).

Table 3
Bis-[6-anilinomethyl-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazol-4-yl] alkanes (**9a–b**)[a]



Compound no.	R	*n*	% Yield	M.p. (°C)	Nature of the compound	Molecular formula	% N found (calc.)
9a[b]	4-Cl	2	74	248–250	light brown micro crystals	$C_{22}H_{18}Cl_2N_{10}S_2$	25.08 (25.2)
9b	3-Cl, 4-F	1	72	110–112	pale yellow micro crystals	$C_{21}H_{14}Cl_2F_2N_{10}S_2$	24.34 (24.2)

[a] Solvent of crystallization, DMF.
[b] IR (KBr): 3334 cm^{-1} (NH), 2924 cm^{-1} (γ_{C-H} ArH), 1593 cm^{-1} ($\gamma_{C=N}$), 1309 cm^{-1} (γ_{C-N}).

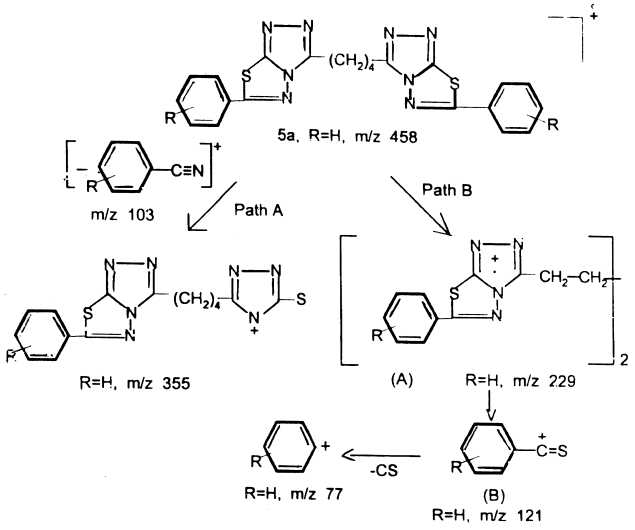
ing to their molecular formula $C_{22}H_{16}C_2N_8O_2S_2$. A peak observed at $m/z = 167$ corresponds to the molecular ion (C). The mass spectral fragmentation of the cyclized product **7b** is given in Scheme 3.

3. Experimental

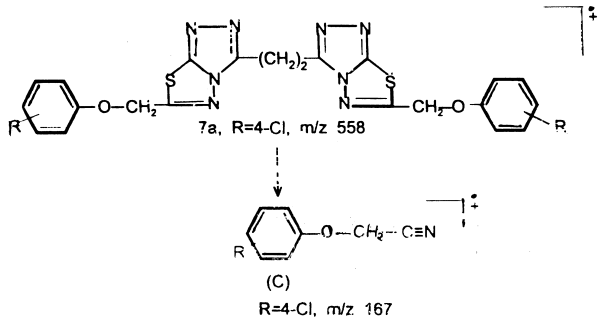
Melting points were taken in open capillary tubes and were uncorrected. IR spectra in KBr pellets were recorded on a Perkin–Elmer 157 IR spectrophotometer. ^{1}H NMR spectra were recorded in DMSO-d_6 on a NMR-EM-390 (90 MHz) spectrometer, using TMS as an internal standard and the mass spectra were recorded on a Jeol JMSD300 spectrometer operating at 70 eV. Purity of the compounds was checked by TLC on silica gel plates using a benzene/methanol (2:1) solvent system and iodine was used as a visualizing agent.

*3.1. Bis-(4-amino-5-mercapto-1,2,4-triazol-3-yl) alkanes (**3a–c**)*

A mixture of dicarboxylic acid (**1**) (0.01 M) and thiocarbohydrazide (**2**) (0.02 M) contained in a flat-bottomed flask was heated in an oil bath until the contents melted. The mixture was maintained at this temperature for 15–20 min (Scheme 1). The product obtained on cooling was treated with dilute sodium bicarbonate solution to remove the unreacted dicarboxylic acid if any. It was then washed with water and collected by filtration. The product was recrystallized from a mixture of dimethylformamide and water to afford the title compounds **3a–c**. **3a**: bis-(4-amino-5-mercapto-1,2,4-triazol-3-yl)methane, m.p. 278–280°C, yield 80%; **3b**: bis-(4-amino-5-mercapto-1,2,4-triazol-3-yl)ethane, m.p. 242–244°C, yield 82%; **3c**: bis-(4-amino-5-mercapto-1,2,4-triazol-3-yl)butane, m.p. 244–246°C, yield 83%.



Scheme 2.



Scheme 3.

3.2. Bis-(6-aryl-1,2,4-triazolo[3,4-b]-1,3,4-triazol-3-yl) alkanes (**5a–h**)

To a mixture of suitable triazolyl alkane (**3**) and aromatic carboxylic acid (**4**), phosphorous oxychloride (10 ml) was added and the contents were heated under reflux for 2 h on a water bath. Excess of phosphorous oxychloride was then distilled off and the residue was poured onto crushed ice while stirring. The resulting solid was washed with water, dilute sodium bicarbonate solution, and then recrystallized from dimethylformamide.

3.3. Bis-(6-aryloxymethyl-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazol-4-yl) alkanes (**7a–f**)

To a mixture of suitable bis-triazolyl alkane (**3**) and aryloxyacetic acid (**6**), phosphorous oxychloride (10 ml) was added and the contents were heated under reflux for 2 h on a water bath. Excess of phosphorous oxychloride was then distilled off and the residue was poured onto crushed ice while stirring. The resulting solid was washed with water, dilute sodium bicarbonate solution and recrystallized from dimethylformamide.

Table 4
Antibacterial activities (minimum inhibitory concentration (μg/ml))

Compound no.	*S. aureus*	*E. coli*	*P. aeruginosa*	*B. subtilis*
5a	12.5	12.5	6	12.5
5c	6	6	6	6
5d	6	6	6	6
5e	3	6	6	3
5g	12.5	6	6	12.5
7e	12.5	6	6	12.5
7b	6	6	6	6
7c	6	6	6	6
7f	3	6	6	3
9a	12.5	6	6	12.5
9b	6	6	6	12.5
Furacin	12.5	6	12.5	12.5

3.4. Bis-(6-anilinomethyl-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazol-4-yl) alkanes (**9a–b**)

To a mixture of suitable bis-triazolyl alkane (**3**) and anilinoacetic acid (**8**), phosphorous oxychloride (10 ml) was added and the contents were heated under reflux for 2 h on a water bath. Excess of phosphorous oxychloride was then distilled off and the residue was poured onto crushed ice while stirring. The resulting solid was washed with water, dilute sodium bicarbonate solution and recrystallized from dimethylformamide.

4. Antibacterial activity

Some of the newly synthesized bis triazolothiadiazolyl alkanes were screened for their in vitro antibacterial activity against *B. subtilis*, *S. aureus*, *P. aeruginosa* and *E. coli* according to the serial dilution method [21]. Solutions of the test compounds were kept in dimethylformamide. Nitrofurazone (Furacin) was used as a standard drug for comparison and solvent control was kept. The minimal inhibitory concentrations (MIC values) of the above compounds are given in Table 4.

5. Results and discussion

All the prepared compounds showed interesting degrees of antibacterial activity. Among the compounds tested, almost all compounds showed higher antibacterial activity than the standard drug. Compounds **5e** and **7f** were the most active and particularly showed very good activity against *B. subtilis*.

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