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Amino Acids in the Syntheses of Heterocyclic Systems: Syntheses and Radiostability of Novel Biologically Active Triazoles Containing the Sulfonamide Moiety

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ABSTRACT: *A number of novel triazoles* **2a–f, 4, 9, 10, 12, 15***; triazolothiadiazoles* **6, 8, 11, 16***; triazolothiadiazine* **5***; and triazolotriazine* **14** *were synthesized and characterized by elemental analyses and spectral data. Six of the compounds showed antifungal activity compared with the fungicide Mycostatine. Radiosterilization of the biologically active compounds* **4, 8, 9b**, *and* **10** *in the dry state may prove to be applicable at the sterile dose 25 kGy.* © 2002 Wiley Periodicals, Inc. Heteroatom Chem 13:316–323, 2002; Published online in Wiley Interscience (www.interscience.wiley.com). DOI 10.1002/hc.10037

INTRODUCTION

The triazole ring system has important and versatile biological activities [1–5]. On the other hand, triazolothiadiazole, thiadiazine, triazine, and sulfonamide derivatives are well known as antifungal agents [6–9]. In view of these findings, we undertook the synthesis of a new series of compounds incorporating the above mentioned biologically active moieties in one molecule and evaluated their antifungal activity. The application of radiation in pharmaceuticals technology has steadily increased during the past few years [10,11]. In the 1997 edition of the European pharmacopoeia, under the methods of preparation of sterile products, irradiation is one of only three processes that can be used as a terminal sterilization method. Also, considerable interest has developed regarding the radiation sensitivity of various antibiotics [12,13] and recently synthesized biologically active compounds [14,15]. Generally, data of these compounds indicate that even at a dose of 25 kGy the radiosterilization may be feasible [16,17].

CHEMISTRY

Several compounds were designed with the aim of exploring their antifungal activity (Schemes 1–4). The starting materials **1a–f** were prepared by reaction of the appropriate sulfonyl chloride derivatives with amino acids in the presence of sodium hydroxide solution (10%), with subsequent acidification [18]. The *s*-triazolosulfonamide derivatives **2a–f** were obtained in good yields by fusion of the

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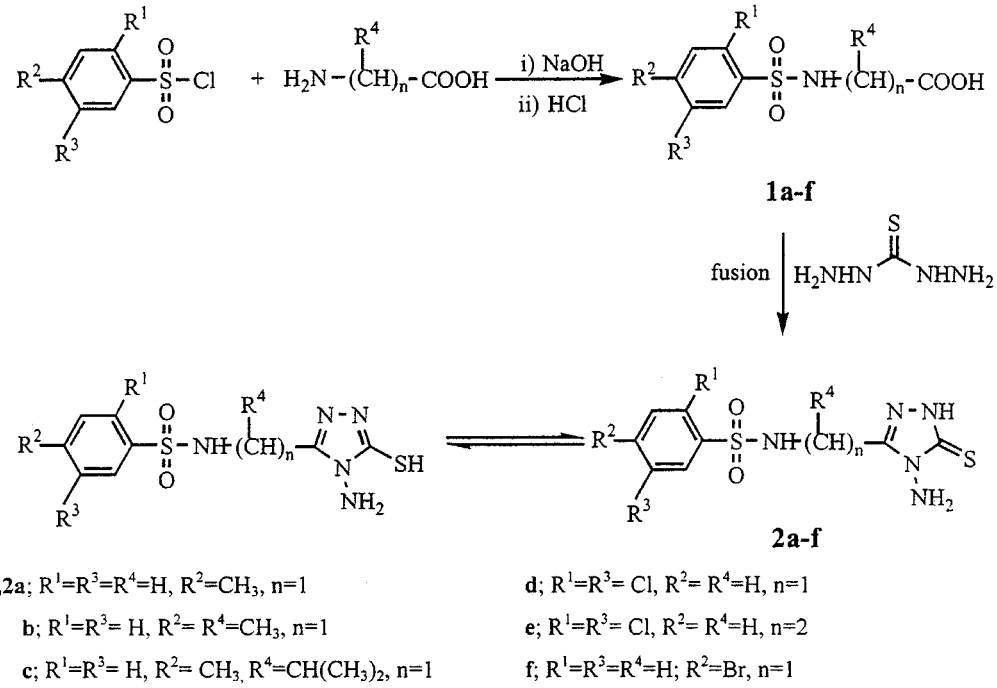


1,2a; $R^1=R^3=R^4=H$, $R^2=CH_3$, n=1
b; $R^1=R^3=H$, $R^2=R^4=CH_3$, n=1
c; $R^1=R^3=H$, $R^2=CH_3$, $R^4=CH(CH_3)_2$, n=1
d; $R^1=R^3=Cl$, $R^2=R^4=H$, n=1
e; $R^1=R^3=Cl$, $R^2=R^4=H$, n=2
f; $R^1=R^3=R^4=H$; $R^2=Br$, n=1

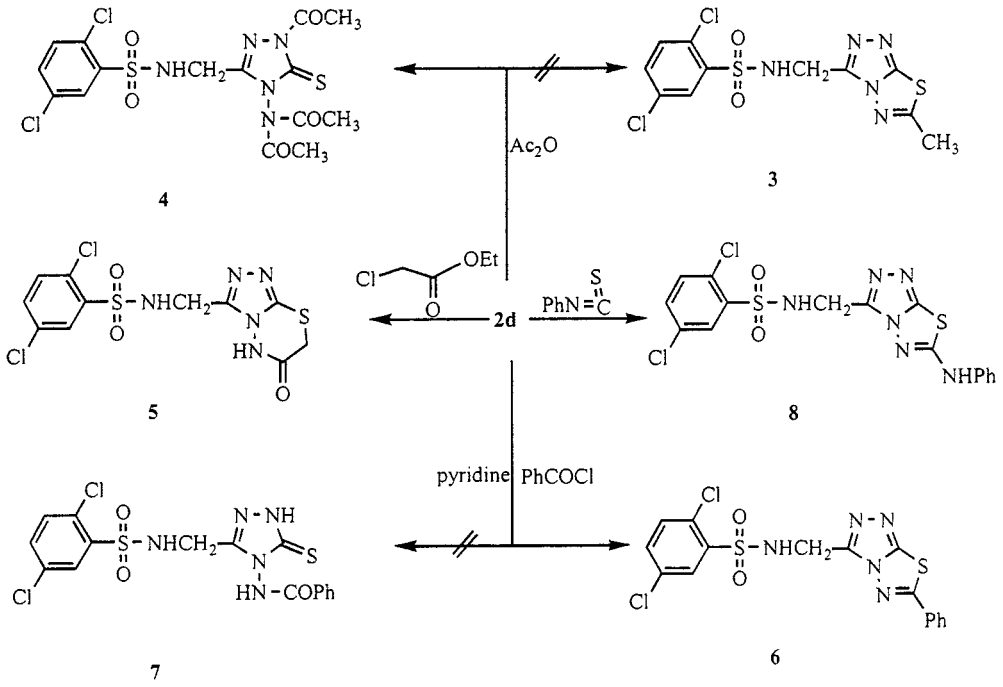
SCHEME 1

respective compounds **1a–f** with thiocarbohydrazide in an oil bath at 180°C (Scheme 1).

The structures of compounds **2a–f** were confirmed on the basis of elemental analyses and spectral data. ^{1}H NMR spectrum of **2a** in DMSO-d_6 exhibited $\delta = 2.4$ (s, 3H, CH_3), 4.0 (d, 2H, CH_2), 5.4 (s, 2H, NH_2), 7.2–7.8 (m, 4H, Ar-H), 8.3 (br, 1H, SO_2NH), 13.4 (s, 1H, NH). MS of **2a** (*m/z*): 284 (M-15, 16.80%), 91 (100%), 149 (15.20%), 105 (13.50%), 59 (20.0%). IR spectrum of **2b** showed bands at 3390, 3330, 3200 (NH, NH_2), 3100 (CH aromatic), 2970 (CH aliphatic), 1650 (C=N), 1320 (C=S). ^{1}H NMR spectrum of **2b** in DMSO-d_6 exhibited $\delta = 1.3$ (d, 3H, CH_3), 2.3 (s, 3H, CH_3 tolyl), 4.5 (q, 1H, CH), 5.4 (s, 2H, NH_2), 7.2–7.8 (m, 4H, Ar-H), 8.6 (br, 1H, SO_2NH), 13.4 (s, 1H, NH). IR spectrum of **2d** showed bands at 3400, 3300, 3150 (NH, NH_2), 3100 (CH aromatic.), 2900 (CH aliphatic), 1590 (C=N). ^{1}H NMR spectrum of **2d** in DMSO-d_6 exhibited $\delta = 4.2$ (d, 2H, CH_2), 5.5 (s, 2H, NH_2), 7.6–8.0 (m, 3H, Ar-H), 8.8 (s, 1H, SO_2NH), 13.6 (s, 1H, NH). MS of **2d** (*m/z*): 353 (M^+, 38.2%), 145 (100%), 209 (34.0%), 163 (17.0%), 144 (72.20), 73 (32.70%), 60 (52.80%). IR spectrum of **2e** showed bands at 3350, 3300, 3200 (NH, NH_2), 3100 (CH aromatic.), 2950 (CH aliphatic), 1620 (C=N). MS of **2f** (*m/z*): 363 (M^+, 27.20%), 144 (100%), 219 (10.30%), 158 (3.90%), 74 (9.60%).

The reaction of compound **2d** with acetic anhydride was carried out in a trial for obtaining triazolothiadiazole derivative **3**, but instead the triacetyl derivative **4** was obtained (Scheme 2). The structure of compound **4** was proved by elemental analyses, IR, and ^{1}H NMR spectroscopy. The IR spectrum showed the absence of NH, NH_2 of the triazole ring and the presence of the SO_2NH moiety at 3150 cm^{-1} and 3C=O groups at 1716, 1690, 1670 cm^{-1}. The ^{1}H NMR spectrum of **4** in DMSO-d_6 revealed $\delta =$ 2.1 (s, 6H, $N(COCH_3)_2$), 2.2 (s, 3H, $NCOCH_3$), 4.8 (s, 2H, CH_2), 7.1–8.0 (m, 3H, Ar-H), 8.9 (s, 1H, SO_2NH). Reaction of **2d** with ethyl chloroacetate furnished the triazolothiadiazine derivative **5**. MS (*m/z*): 395 (M^+, 35.71%), 180 (100%), 355 (35.71%), 285 (64.29%), 220 (64.29%), 176 (39.29%), 118 (32.14%), 94 (39.29%), 78 (3.57%). Also, reaction of **2d** with benzoyl chloride in pyridine effected cyclization to furnish triazolothiadiazole **6**, rather than the benzoylaminotriazole derivative **7**, based on the elemental analyses and IR spectrum, which showed the absence of a C=O band and the presence of an NH band at 3460, a CH aliphatic group at 2910, and a C=N group at 1610 cm^{-1}. MS of **6** (*m/z*): 439 (M^+, 1.52%), 73 (100%), 355 (4.66 %), 275 (10.18%), 207 (46.43%), 150 (30.35%), 107 (24.64%), 60 (68.13%).

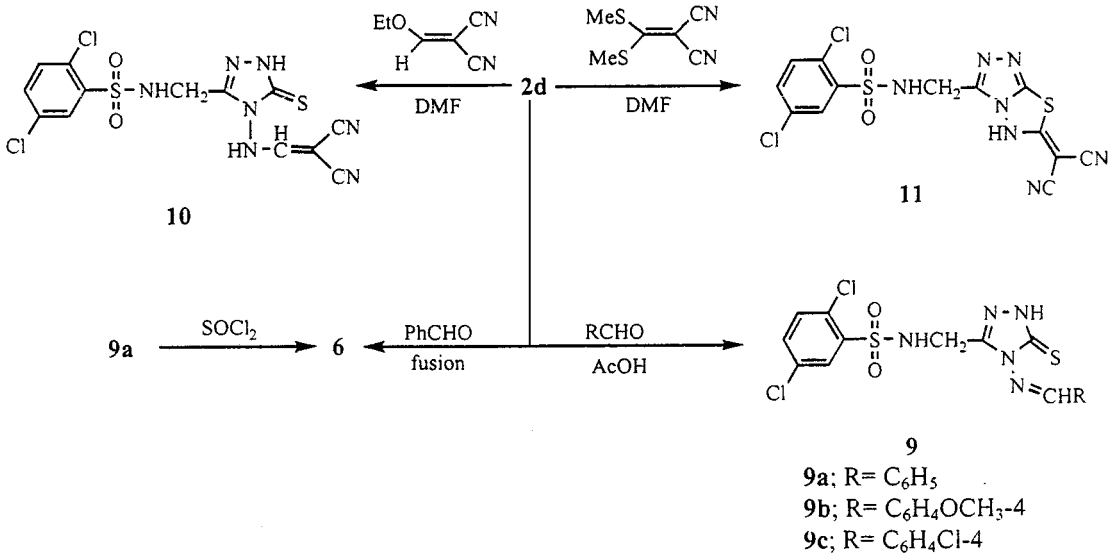
The triazolothiadiazole derivative **8** was obtained in good yield by the reaction of **2d** with phenyl isothiocyanate in pyridine. The structure of **8** was established by elemental analyses and its IR spectrum, which showed the absence of NH_2 bands and presence of NH at 3150 cm^{-1}, and of C=N at 1595 cm^{-1}.



SCHEME 2

Condensation of **2d** with aromatic aldehydes in glacial acetic acid afforded Schiff bases **9a–c**, while, under condition of fusion, the reaction of **2d** with benzaldehyde caused cyclization to afford the corresponding triazolothiadiazole derivative **6**. The structure of **6** was also confirmed through its synthesis by reaction of **9a** with thionyl chloride (Scheme 3). The ^{1}H NMR spectrum of **9b** in DMSO-d_6 revealed $\delta = 3.8$ (s, 3H, OCH_3), 4.3 (s, 2H, CH_2), 7.0–7.9 (m, 7H, Ar-H), 8.3 (s, 1H, SO_2NH), 9.7 (s, 1H, N=CH), 13.9 (s, 1H, NH).

The dicyano derivatives **10** and **11** were obtained in good yields by reaction of compound **2d** with ethoxymethylenemalononitrile and [bis(methylsulfanyl)methylidene]malononitrile, respectively in dimethylformamide. The IR spectrum of **10** showed bands at 3430 (NH), 2925 (CH aliphatic), 2211 (C≡N), 1615 (C=N). MS of **10** (*m/z*): 429 (M^+, 0.67%), 55 (100%), 368 (4.88%), 327 (3.33%), 257 (8.86%), 152 (17.44%), 111 (41.07%), 73 (53.57%). The IR spectrum of **11** showed bands at 3200 (NH), 3100 (CH aromatic.), 2927 (CH



SCHEME 3

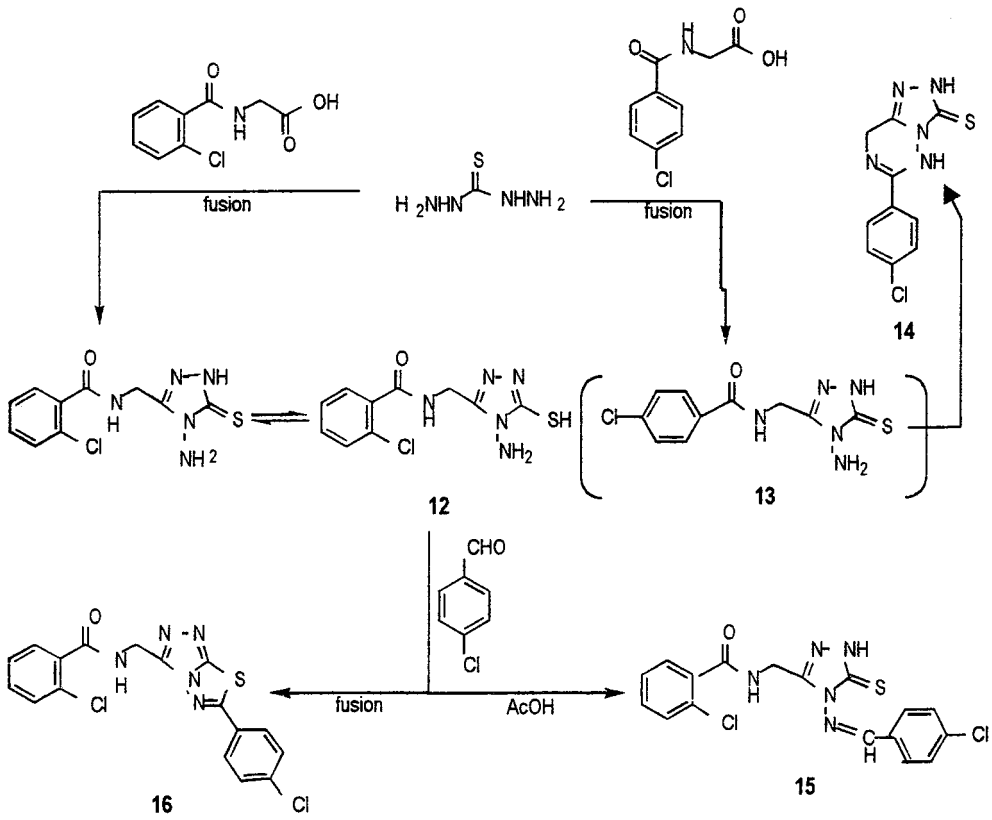
aliphatic), 2210 (C≡N), 1620 cm^{-1} (C=N). MS of **11** (*m/z*): 427 (M^+, 5.22%), 177 (100%), 386 (18.26%), 354 (36.09%), 284 (48.26%), 193 (37.39%), 148 (15.22%), 106 (80.43%), 62 (45.65%).

Fusion of each thiocarbohydrazide with 2-chlorohippuric acid afforded the corresponding triazole derivative **12**, while, with 4-chlorohippuric acid, double cyclization occurred to give the triazolotriazine **14** through the formation of the expected triazole derivative **13** (Scheme 4). The structures of **12** and **14** were established by elemental analyses and by IR, ^1H NMR, and MS spectroscopy. The IR spectrum of **12** showed bands at 3328, 3250, 3182 (NH, NH_2), 3087 (CH aromatic.), 2947 (CH aliphatic), 1640 (C=O), 1592 (C=N). The ^1H NMR spectrum of **12** in DMSO-d_6 exhibited $\delta = 4.5$ (d, 2H, CH_2), 5.6 (s, 2H, NH_2), 7.4–7.6 (m, 4H, Ar-H), 8.9 (t, 1H, CONH), 9.0 (s, 1H, NH triazole). The IR spectrum of **14** showed the absence of (C=O) and the presence of NH at 3250, the CH aromatic at 3100, the C=N group at 1620, 1595 cm^{-1}. MS of **14** (*m/z*): 265 (M^+, 0.1%), 131 (100%), 236 (0.7%), 192 (0.1%), 158 (0.31%), 116 (0.74%), 85 (7.76%), 60 (11.25%).

Condensation of **12** with 4-chlorobenzaldehyde in acetic acid furnished the Schiff base **15**, while, under the conditions of fusion, the reaction of **12** with 4-chlorobenzaldehyde effected cyclization to give the corresponding triazolothiadiazole **16**. IR spectrum of compound **15** exhibited bands at 3276 (NH), 3094 (CH aromatic.), 2942 (CH aliphatic), 1640 (C=O), 1590 (C=N). The ^1H NMR of **15** in DMSO-d_6 revealed $\delta = 4.6$ (d, 2H, CH_2), 7.2–8.0 (m, 8H, Ar-H), 9.0 (t, 1H, CONH), 10.1 (s, 1H, N=CH), 14.0 (s, 1H, NH triazole). The ^1H NMR of compound **16** in DMSO-d_6 showed $\delta = 4.5$ (d, 2H, CH_2), 7.3–8.1 (m, 8H, Ar-H), 9.0 (t, 1H, CONH).

EXPERIMENTAL

All melting points are uncorrected and were determined on an electrothermal STUART melting point apparatus (SCIENTIFIC Co. Ltd, UK). IR spectra (cm^{-1}) were recorded on a Pye-Unicam spectrophotometer, type 1200, using the KBr technique. ^1H NMR spectra were recorded on a Varian EM-390 (90 MHz) spectrometer using TMS as an internal standard, DMSO-d_6 as a solvent and chemical shifts were expressed in δ values. Mass spectra were run using an HP Model: MS-5988 instrument. Elemental analyses were determined using a Perkin-Elmer 240 (Microanalyses) instrument. The samples were irradiated with gamma radiation (^{60}Co) at the National Center for Radiation Research and Technology. Powder samples contained in polycarbonate vials were



SCHEME 4

irradiated at room temperature. UV spectra were recorded using a ATI Unicam UV–VIS Aurora Scan instrument.

*General Procedure for Syntheses of Tosyl Amino Acids (**1a–f**)*

The tosyl amino acid derivatives were prepared according to the procedure of McChesney et al. [18], where each amino acid (0.026 mol) was dissolved in 1 N NaOH (25 ml) and in a period of 13 min a solution of *p*-toluenesulphonyl chloride or 2,5-dichlorophenylsulphonyl chloride or *p*-bromophenylsulphonyl chloride (0.027 mol) in ether (30 ml) was added in portions. The mixture was stirred at room temperature for 3 h. The excess sulphonyl chloride was filtered off and the solution treated with 2 N HCl until acidic to congo red indicator (pH 5). After cooling, acidification caused the product to precipitate. The crude product was filtered off, washed with water, and dried. The crude materials were recrystallized to give **1a–f**.

*General Procedure for Syntheses of s-Triazolosulfonamide Derivatives (**2a–f**)*

A mixture of thiocarbohydrazide (1.06 g, 0.01 mol) and different sulfonyl amino acids **1a–f** (0.01 mol) were fused at 180°C in an oil bath for 15 min. After cooling, each reaction mixture was triturated with ethanol to give (4-amino-5-mercapto-*s*-triazol-3-yl)-methyl-4-tolylsulfonamide (**2a**), (4-amino-5-mercapto-*s*-triazol-3-yl)-methyl-ethyl-4-tolylsulfonamide (**2b**), (4-amino-5-mercapto-*s*-triazol-3-yl)-methyl-isobutyl-4-tolylsulfonamide (**2c**), (4-amino-5-mercapto-*s*-triazol-3-yl)-methyl-methyl-2,5-dichlorobenzenesulfonamide (**2d**), (4-amino-5-mercapto-*s*-triazol-3-yl)-methyl-ethyl-2,5-dichlorobenzenesulfonamide (**2e**), and (4-amino-5-mercapto-*s*-triazol-3-yl)-dimethyl-4-bromobenzenesulfonamide (**2f**).

*(1-Acetyl-4-diacetylamino-5-thioxo-s-triazol-3-yl)-methyl-2,5-dichlorobenzenesulfonamide (**4**)*

A solution of **2d** (3.5 g, 0.01 mol) in acetic anhydride (20 ml) was heated under reflux for 10 h. After cooling the excess of acetic anhydride was removed under reduced pressure. The obtained solid was recrystallized from acetic acid to give **4**.

*(6-Oxo-5H-s-triazolo[3,4-b][1,3,4]thiadiazin-3-yl)-methyl-2,5-dichlorobenzenesulfonamide (**5**)*

To a solution of **2d** (3.5 g, 0.01 mol) in dioxane (50 ml) and triethylamine (1.01 g, 0.01 mol), ethyl chloroacetate (1.22 g, 0.01 mol) was added. The reaction mixture was refluxed for 2 h. The formed product was filtered off, washed with water, dried and recrystallized from ethanol to give **5**.

*(6-Phenyl-s-triazolo[3,4-b][1,3,4]thiadiazol-3-yl)-methyl-2,5-dichlorobenzenesulfonamide (**6**)*

Method A: A solution of compound **2d** (3.5 g, 0.01 mol) in dry pyridine (20 ml) and benzoyl chloride (1.40 g, 0.01 mol) was added. The reaction mixture was refluxed for 8 h. After cooling and acidification with dil. HCl, the precipitate was formed and then collected by filtration, washed with water, and recrystallized from dioxane to give **6**.

Method B: A mixture of **2d** (3.5 g, 0.01 mol) and benzaldehyde (1.06 g, 0.01 mol) was fused for 15 min at 220°C in an oil bath. After cooling, the reaction mixture was triturated with ethanol to give **6**.

Method C: A solution of **9a** (4.41 g, 0.01 mol) in thionyl chloride (10 ml) was refluxed for 1 h and the obtained solid was recrystallized from dioxane to give **6**.

*(Anilino-s-triazolo[3,4-b][1,3,4]thiadiazol-3-yl)-methyl-2,5-dichlorobenzenesulfonamide (**8**)*

A solution of **2d** (3.5 g, 0.01 mol) and phenyl isothiocyanate (1.35 g, 0.01 mol) in dry pyridine (20 ml) was refluxed until the evolution of H_2S had ceased (12 h). The reaction mixture was poured into ice-cold water (100 ml). The precipitate was filtered off, dried and recrystallized from ethanol to give **8**.

*(4-Arylideneamino-1H-5-thioxo-s-triazol-3-yl)-methyl-2,5-dichlorobenzenesulfonamides (**9a–c**)*

A mixture of **2d** (3.5 g, 0.01 mol) and benzaldehyde or 4-methoxybenzaldehyde or 4-chlorobenzaldehyde in acetic acid (10 ml) was refluxed for 5 h. The solvent was evaporated under reduced pressure. The solid product was collected and recrystallized from acetic acid to give **9a–c**.

*(4-Aminomethylenemalononitrile-1H-5-thioxo-s-triazol-3-yl)methyl-2,5-dichlorobenzenesulfonamide (**10**) and (6-dicyanoethylidene-5H-s-triazolo[3,4-b]thiadiazol-3-yl]methyl-2,5-dichlorobenzenesulfonamide (**11**)*

A mixture of **2d** (3.5 g, 0.01 mol), ethoxymethylenemalononitrile or [bis(methylsulphonyl)methylidine] malononitrile (0.01 mol), and triethylamine (1.01 g, 0.01 mol) in dimethylformamide (20 ml) was

TABLE 1 Characterization Data for Newly Synthesized Compounds

				Elemental Analysis [Calculated/Found (%)]		
Compound	*M.P. (°C)*	*Yield (%)*	*Mol. Formula (Mol. Wt)*	*C*	*H*	*N*
2a	200–202	80	$C_{10}H_{13}N_5O_2S_2$ (299)	40.13 40.00	4.35 4.50	23.41 23.20
2b	195–197	85	$C_{11}H_{15}N_5O_2S_2$ (313)	42.17 42.40	4.79 4.60	22.36 22.20
2c	218–220	79	$C_{13}H_{19}N_5O_2S_2$ (341)	45.75 45.55	5.57 5.50	20.53 20.70
2d	205–207	74	$C_9H_9N_5O_2S_2Cl_2$ (353)	30.59 30.80	2.55 2.50	19.83 19.60
2f	210–212	70	$C_9H_{10}N_5O_2S_2Br$ (363)	29.75 29.60	2.75 2.55	19.28 19.40
2e	214–216	76	$C_{10}H_{11}N_5O_2S_2Cl_2$ (367)	32.69 32.80	2.99 2.80	19.07 19.00
4	70–72	75	$C_{15}H_{15}N_5O_5S_2Cl_2$ (479)	37.57 37.80	3.13 3.40	14.61 14.30
5	150–152	72	$C_{11}H_9N_5O_3S_2Cl_2$ (393)	33.58 33.30	2.29 2.60	17.81 17.50
6	>300	72	$C_{16}H_{11}N_5O_2S_2Cl_2$ (439)	43.74 43.40	2.51 2.70	15.95 16.20
8	90–92	79	$C_{16}H_{12}N_6O_2S_2Cl_2$ (454)	42.29 42.50	2.64 2.50	18.50 18.80
9a	260–262	79	$C_{16}H_{13}N_5O_2S_2Cl_2$ (441)	43.53 43.70	2.95 2.80	15.87 15.90
9b	180–182	82	$C_{17}H_{15}N_5O_3S_2Cl_2$ (471)	43.31 43.60	3.18 3.40	14.86 15.10
9c	290–292	88	$C_{16}H_{12}N_5O_2S_2Cl_3$ (475)	40.42 40.60	2.53 2.60	14.74 14.60
10	155–157	83	$C_{13}H_9N_7O_2S_2Cl_2$ (429)	36.36 36.00	2.09 2.30	22.84 22.60
11	120–122	81	$C_{13}H_7N_7O_2S_2Cl_2$ (427)	36.53 36.20	1.63 1.40	22.95 22.70
12	210–212	70	$C_{10}H_{10}N_5OSCl$ (283)	42.40 42.20	3.53 3.10	24.73 24.90
13	205–207	78	$C_{10}H_8N_5SCl$ (265)	45.28 45.00	3.02 3.20	26.42 26.20
15	253–255	76	$C_{17}H_{13}N_5OSCl_2$ (405)	50.37 50.10	3.20 3.50	17.28 17.40
16	280–282	56	$C_{17}H_{13}N_5OSCl_2$ (405)	50.37 50.50	3.20 3.40	17.28 17.60

refluxed for 10 h. The obtained solid was recrystallized from dioxane to give **10** and **11**, respectively.

*(4-Amino-5-mercapto-s-triazol-3-yl)methyl-2-chlorobenzenecarboxamide (***12***) and 6-(4′-chlorophenyl)-1,4,7-trihydro-9-thioxo-s-triazolo-[3,4-b][1,2,4]triazine (***14***)*

A mixture of thiocarbohydrazide (1.06 g, 0.01 mol) and 2-chlorohippuric acid or 4-chlorohippuric acid (2.13 g, 0.01 mol) was fused at 180°C in an oil bath for 15 min. After cooling, the reaction mixture was triturated with ethanol and the obtained solid was recrystallized from ethanol to give **12** and **14**, respectively.

*[4-(4′-Chlorobenzylideneamino)-1H-5-thioxo-s-triazol-3-yl]methyl-2-chlorobenzenecarboxamide (***15***)*

A mixture of **12** (2.83 g, 0.01 mol) and 4-chlorobenzaldehyde (1.40 g, 0.01 mol) in acetic acid (20 ml) was refluxed for 4 h. The solvent was evaporated under reduced pressure. The solid product was recrystallized from acetic acid to give **15**.

*[6-(4-Chlorophenyl)-s-triazolo[3,4-b][1,3,4]thiadiazol-3-yl)methyl-2-chlorobenzenecarboxamide (***16***)*

A mixture of **12** (2.83 g, 0.01 mol) and 4-chlorobenzaldehyde (1.40 g, 0.01 mol) was fused at

TABLE 2 Antifungal Activity of Some Newly Synthesized Compounds (Inhibition Zones, mm)

	Aspergillus ochraceus Wilhelm (AUCC-230)	*Aspergillus flavus Link (AUCC-164)*	*Penicillium chrysogenum Thom (AUCC-530)*	*Candida albicans (Robin) Berkho (AUCC-1720)*
2a	18	18	20	10
2b	24	24	24	24
2c	22	20	20	18
2d	20	22	20	20
2f	34	36	40	40
4	32	34	40	40
6	40	40	40	34
8	24	22	34	35
9b	40	40	35	34
10	34	32	30	24
Mycostatine[a]	40	40	42	40

[a]Manufactured by Bristol-Myers Squibb, Giza, Egypt.

220°C in an oil bath for 15 min. After cooling, the reaction mixture was triturated with ethanol to give **16**.

ANTIFUNGAL ACTIVITY

Most of the newly synthesized compounds were screened for their antifungal activity against four species of fungi, namely *Aspergillus ochraceus* Wilhelm (AUCC-230), *Aspergillus flavus* Link (AUCC-164); *Penicillium chrysogenum* Thom (AUCC-530), and *Candida albicans* (Robin) Berkho (AUCC-1720), using a cup plate agar diffusion method [19]. The fungi cultures were maintained on Czapek's Dox agar medium. The tested compounds were dissolved in *N*,*N*-dimethylformamide (DMF) to get a solution of 1 mg/ml concentration. The inhibition zones were measured in millimeters at the end of an incubation period of 48 h at 28°C. Dimethylformamide showed no inhibition zones. Mycostatine was used as a standard reference fungicide to evaluate the potency of the tested compounds. The minimal inhibitory concentration (MIC) of the active compounds was measured using the serial dilution method [20].

The results are illustrated in Table 2. The antifungal activity of the synthesized compounds showed that the halogenated triazoles having sulfonamide moieties **2f, 4, 6, 8, 9b**, and**10** were found to be the most active compounds IZ (24–40 mm) against all the fungi under investigation, (MIC values were 100 μg/ml). These results indicate that the biologically active compounds **2f, 4, 6, 8, 9b**, and **10** are nearly as active as the standard Mycostatine (30 μg/ml).

RADIOSTABILITY OF THE BIOLOGICALLY ACTIVE COMPOUNDS

The aim of the present work is to investigate the stability of the chemical structure of the biologically active compounds **4, 8, 9b**, and **10** before sterilization. These compounds were irradiated in the dry state (doses of gamma irradiation ranging from 5–40 kGy) at dose rate 1 kGy/7 min. Ultraviolet spectra

TABLE 3 UV Spectra of Biologically Active Compounds 4, 8, 9b, and 10 in 10^{-4} M DMF Solution Before and After γ-Irradiation

Compound	*Dose (kGy)*	λ_{max} *(1)*	*Abs./O.D.*[a]
4	0[b]	275	0.460
	5		0.471
	10		0.481
	15		0.574
	20		0.579
	25		0.630
	30		0.657
	40		0.657
8	0[b]	285	1.272
	5		1.421
	10		1.433
	15		1.544
	20		1.624
	25		2.044
	30		2.173
	40		2.249
9b	0[b]	290	1.373
	5		1.566
	10		1.633
	15		1.648
	20		1.652
	25		1.673
	30		1.748
	40		1.811
10	0[b]	280	0.926
	5		1.208
	10		1.335
	15		1.342
	20		1.349
	25		1.349
	30		1.629
	40		1.640

[a]Abs./O.D. = absorbance/optical density.
[b]Nonirradiated compound (control).

of nonirradiated (control) and irradiated compounds in DMF as solvent are listed in Table 3.

The results showed that all the biologically active compounds **4, 8, 9b**, and **10** were radioresistant, retaining their structure unchanged up to 40 kGy (the absorbance value above control). Also, thin layer chromatographic analyses for compounds **4, 8, 9b**, and **10** was conducted before and after irradiation using precoated silica gel G sheet 1B-F and a mixture of 2:1 ethylacetate–petroleum ether as eluent. Spots were detected by use of a UV lamp at 254 nm. Compounds **4, 8, 9b**, and **10** showed a single distinct spot, before and after irradiation, with the same R_f values of 0.42, 0.81, 0.94, and 0.90, respectively. This means that no change occurred in the structures of these compounds, so the radiosterilization of these compounds in the dry form may prove to be applicable.

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