Synthesis and Fungicidal Activity of Some New of 2,4-Dichlorobenzoic acid -5-sulphonyl Amino Acid Derivatives

RAGAB A. EL-SAYED*, N.S. KHALAF, F.A. KORA AND M.F. BADIE
Chemistry Department, Faculty of Science, Al-Azhar University Nasr-City, Cairo, Egypt

(Received 30th January, 1991)

Summary: 2,4-Dichlorobenzoic acid reacts with chlorosulphonic acid to yield the corresponding sulphonyl chloride (1). Subsequent condensation with nucleophiles afforded sulphonyl derivatives (2-11), which are used for the synthesis of methyl esters (12-18), the corresponding hydrazide (19-22) and the dipeptide derivatives (23-36). The spectral data of the synthesized compounds (2-36) and the results of preliminary biological screeing are briefly discussed

Introduction

The work reported here is a continuation of our programme on the synthesis and reactivity of sulphonyl derivatives as candidate pesticides [1-4], and extends on previous chlorosulphonation of carboxylic acids [5-7].

Many sulphonyl derivatives such as amides [8], azides [9], and hydrazides [10-12] have valuable biocidal properties, for instance, as antibacterials, nematicides, and fungicides.

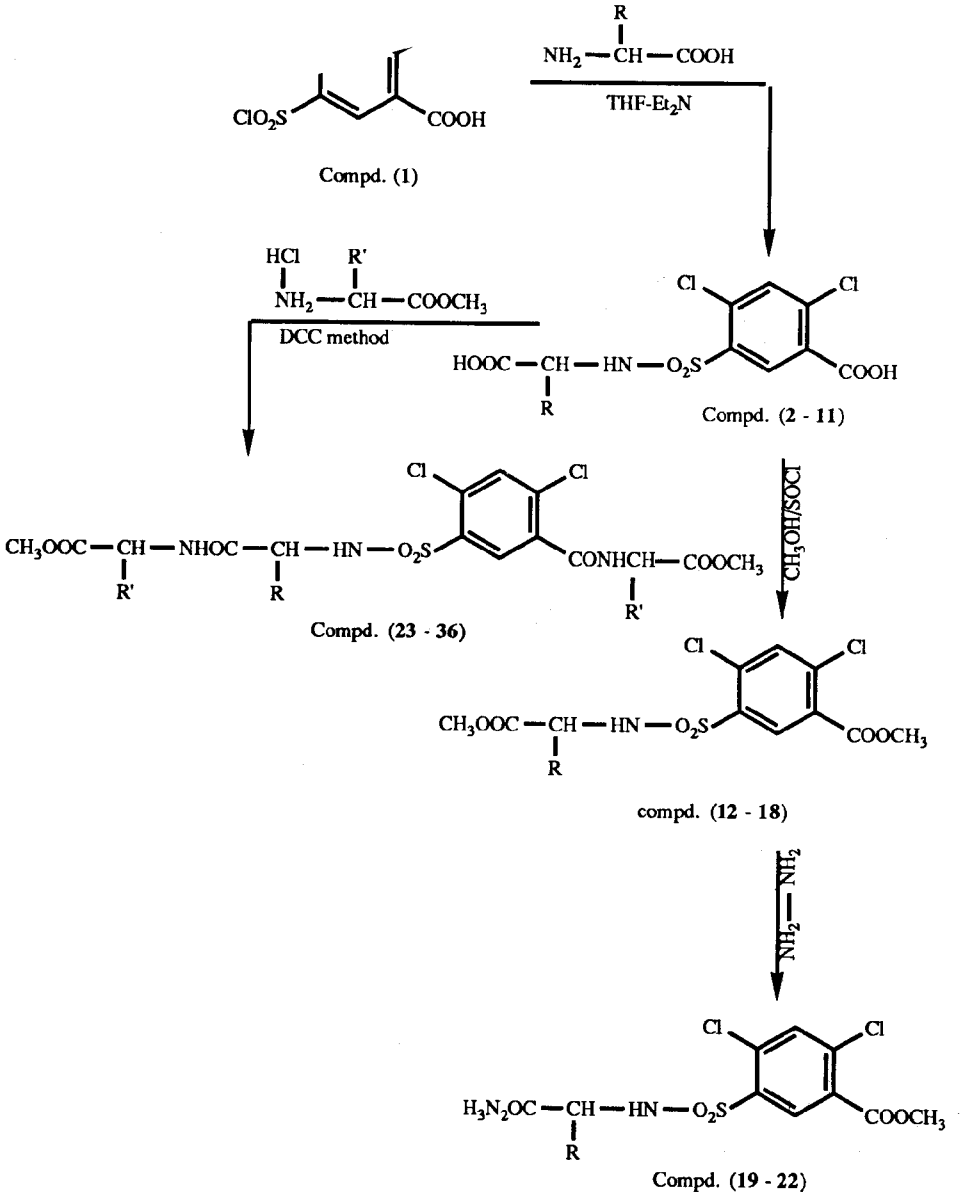
We have studied the chlorosulphonation of 2,4-dichlorobenzoic acid to obtain a range of novel sulphonyl derivatives for biocidal evaluation.

2,4-Dichlorobenzoic acid reacts with chlorosulphonic acid to give an excellent yield of the corresponding sulphonyl chloride (1) according to the procedure described earlier [13]. Reaction of (1) with different amino acids using tetrahydrofuran - triethylamine afforded the sulphonamide derivative (2-11) (Scheme 1). Treatment of these derivatives (2-11) with absolute methanol and pure thionyl chloride, at 0°C, yielded methyl esters (12-18), which upon treatment with hydrazine hydrate (85%) for 24 hrs, at room temperature, gave the sulphonyl hydrazides (19-22) described in Table 1.

Reaction of the same sulphonamides (2-11) with 2 moles of amino acid methyl ester hydrochlorides in tetrahydrofuran, using carbodiimide (DCC) method afforded the dipeptide derivatives (23- 36) (Scheme 1).

All the compounds synthesized (2-36) were characterized by micro- analysis, spot test, and spectroscopic data Table 1.

*To whom all correspondance should be addressed.



Scheme 1

The IR spectra exhibited the normal NH; C=O and CONH absorption [14] and the PMR spectra showed characteristic absorptions confirming the assigned structures. Antibacterial screening of the synthesized compounds (2-36) was carried out using the hole plate method and filter paper disc method [15-17].

All the synthesized compounds were tested against gram-positive, gram-negative, and fungi. The microorganisms tested included *Bacillus cereus* (NRRL-B-569), *Bacillus sphaericus* (159), *Staphylococcus aureus* (ATCC-6538P), *Sarcino* species and *Escherichia coli* NRRL-B-210.

A qualitative screen was performed on all compounds while quantitative assays were done on active compounds only. Several of the sulphonyl amino acids (4) (6), (7), (10) (11) and some of the corresponding dipeptide methyl esters (27), (31) (35), (36) gave complete control of the bacteria with MIC of 10-100 μg/ml (cf. Table 2). The other

Table 1: Physical data of various 2,4-dichlorobenzoic acid-5-sulphonyl amino acids, esters, hydrazides, and dipeptide derivatives (2-36)

Com-pound	-R	Yield	M.p.°C	Rf	cryst.	$[\alpha]D^{20}$ solvent	Mol.formula	Elemental analysis % C	H	N	C	H	N
								Calc.			Found		
2	Gly	54	200-202	0.60	a	-	$C_9H_7NO_6Cl_2S$	32.93	2.13	4.27	32.90	2.11	4.25
3	DL-Ala	46	219-220	0.73	a	-	$C_{10}H_9N)_6O_2S$	35.09	2.63	4.09	35.00	2.55	4.03
4	B-Ala	51	230-232	0.55	a	-	$C_{10}H_9NO_6Cl_2S$	35.09	2.63	4.09 3	5.04	2.60	4.00
5	L-Val	65	198-200	0.58	a	+75.36	$C_{12}H_{13}NO_6Cl_2S$	38.92	3.51	3.78	38.87	3.46	3.77
6	L-Leu	56	210-212	0.67	a	+128.11	$C_{13}H_{15}NO_6Cl_2S$	40.62	3.91	40.55	3.88	3.61	
7	DL-Leu	62	170-172	0.80	a	-	$C_{13}H_{15}NO_6Cl_2S$	40.62	3.91	3.65	40.59	3.91	3.59
8	L-Phe	50	178.180	0.62	a	+50.24	$C_{16}H_{13}NO_6Cl_2S$	45.93	3.11	3.35	45.91	3.00	3.35
9	DL-Phe	53	145.147	0.64	a	-	$C_{16}H_{13}NO_6Cl_2S$	45.93	3.11	3.35	45.90	3.09	3.22
10	L-Tyr	56	118-120	0.50	a	+175.84	$C_{16}H_{13}NO_7Cl_2S$	44.24	3.00	3.23	44.13	3.00	3.11
11	L-Asp	44	138-140	0.66	a	+66.14	$C_{11}H_9NO_8Cl_2S$	34.20	2.33	3.63	34.16	2.31	3.59
12	Gly-OMe	53	118-120	0.72	b	-	$C_{11}H_{11}NO_6Cl_2S$	37.08	3.09	3.93	37.01	3.06	3.91
13	DL-Ala-OMe	62	140-142	0.70	b	-	$C_{12}H_{13}NO_6Cl_2S$	38.92	3.51	3.78	38.87	3.50	3.78
14	B-Ala-OMe	73	103-105	0.73	b	-	$C_{12}H_{13}NO_6Cl_2S$	38.92	3.51	3.78	38.91	3.50	3.71
'5	L-Val-OMe	79	120-122	0.69	b	+52.75	$C_{14}H_{17}NO_6Cl_2S$	42.21	4.27	3.52	42.16	4.22	3.49
16	L-Leu-OMe	66	96-98	0.63	b	+105.50	$C_{15}H_{19}NO_6Cl_2S$	43.69	4.61	3.40	43.56	4.53	3.38
17	DL-Leu-OMe	56	105-107	0.66	b	-	$C_{15}H_{19}NO_6Cl_2S$	43.69	4.61	3.40	43.61	4.58	3.39
18	L-Phe-OMe	55	88-90	0.61	b	+70.33	$C_{18}H_{17}NO_6Cl_2S$	48.43	3.81	3.14	48.41	3.78	3.11
19	Gly-N_2H_3	85	110-112	0.44	c	-	$C_9H_{11}N_5O_4Cl_2S$	30.34	3.09	19.66	30.39	3.11	19.89
20	DL-Ala-N_2H_3	78	190-192	0.36	c	-	$C_{10}H_{13}N_5O_4Cl_2S$	32.43	3.51	18.92	32.4	3.47	18.96
21	B-Ala-N_2H_3	73	90-92	0.42	c	-	$C_{10}H_{13}N_5O_4Cl_2S$	32.43	3.51	18.92	32.4	1 3.50	18.91
22	L-Leu-N_2H_3	79	75-77	0.47	c	+90.43	$C_{13}H_{19}N_5O_4Cl_2S$	37.66	4.61	17.00	37.83	4.59	17.00
23	Gly-Gly-OMe	62	185-187	0.57	a	-	$C_{15}H_{17}N_3O_8Cl_2S$	38.30	3.62	8.94	38.30	3.61	8.91
24	Dl-Ala-Gly-OMe	65	193-195	0.52	a	-	$C_{16}H_{19}N_3O_8Cl_2S$	39.67	3.93	8.68	39.64	3.90	8.61
25	L-Val-Gly-OMe	66	178-180	0.48	a	+85.40	$C_{18}H_{23}N_3O_8Cl_2S$	42.19	4.49	8.20	42.14	4.44	8.16
26	L-Leu-Gly-OMe	72	163-165	0.56	a	+110.52	$C_{19}H_{25}N_3O_8Cl_2S$	43.35	4.75	7.98	43.32	4.69	7.91
27	L-Phe-Gly-OMe	81	170-172	0.44	a	+105.50	$C_{22}H_{23}N_3O_8Cl_2S$	47.14	4.11	7.50	47.11	4.04	7.47
28	Gly-DL-Ala-OMe	85	165-167	0.65	a	-	$C_{17}H_{21}N_3O_8Cl_2S$	40.96	4.22	8.43	40.16	4.11	8.41
29	DL-Ala-DL-Aal-OMe	60	143-145	0.58	a	-	$C_{18}H_{23}N_3O_8Cl_2S$	42.19	4.49	8.20	42.13	4.42	8.16
30	L-Leu-DL-Ala-OMe	63	152-154	0.44	a	+120.57	$C_{21}H_{30}N_3O_8Cl_2S$	45.41	5.41	7.57	45.31	5.32	7.57
31	L-Phe-DL-Ala-OMe	76	166-168	0.53	a	+140.67	$C_{24}H_{27}N_3O_8Cl_2S$	48.98	4.59	7.14	48.99	4.60	7.13
32	Gly-L-Leu-OMe	50	64-68	0.51	a	+100.48	$C_{23}H_{33}N_3O_8Cl_2S$	47.42	5.67	7.22	47.40	5.66	7.16
33	DL-Ala-L-Leu-OMe	53	140-142	0.58	a	+145.69	$C_{24}H_{35}N_3O_8Cl_2S$	48.32	5.87	7.05	48.22	5.86	7.05
34	L-Val-L-Leu-OMe	50	165-167	0.61	a	+165.79	$C_{26}H_{39}N_3O_8Cl_2S$	50.00	6.25	6.73	50.00	6.13	6.69
35	L-Leu-L-Leu-OMe	63	176-178	0.41	a	+135.64	$C_{26}H_{40}N_3O_8Cl_2S$	49.92	6.40	6.72	49.88	6.33	6.66
36	L-Phe-L-Leu-OMe	50	174-176	0.45	a	+130.62	$C_{30}H_{39}N_3O_8Cl_2S$	53.57	5.80	6.25	53.49	5.76	6.19

*Crystallization solvents a = methanol-water, b = methanol-ether, c = ethanol-water
(2-11), R' = OH ; (23-27), R' = Gly-OMe
(12-18), R' = OCH_3; (28-31), R' = DL-Ala-OMe
(19-22), R' = N2H3; (32-36, R' = L-Leu-OMe

compounds i.e. the methyl ester (12-18) and the corresponding hydrazides (19-22) were found to be inactive towards the tested microorganisms.

Other pharmacological investigations are currently in progress.

Experimental

Melting points were determined using electrothermal melting point apparatus and are uncorrected. Thin layer chromatography (R_f value) for analytical purposes was carried out on silica gel G1 plastic sheets and developed with n-butanol:acetic acid:water (4:1:1) using iodine, ninhydrin, and benzidine as spraying agents. Optical rotation $[\alpha]D^{20}$ were measured for all compounds in DMF at λ_{max} 589 n.m on Bellingham stanely polarimeter using 5 cm tube at 20°C. The infrared spectra (ν_{max}, cm^{-1}) were taken in KBr disc using Schimadzu IR - 408, instrument, PMR spectra (chemical shifts δ in ppm) were measured in

Table 2: Minimal inhibitory concentration (MIC) in μg/ml of the biologically active compounds

Compd. No.	Staph. aureus	Sarc. species	Bac. cereus	Bac. sphae.	Eschi coli
4	25	25	-	50	50
6	10	10	50	50	50
7	-	50	50	100	25
10	50	50	-	-	-
11	-	25	50	25	50
27	-	10	-	25	50
31	100	100	100	100	100
35	10	50	50	100	100
36	25	50	50	50	50

DMSO-d_6 using Varian EM- 360 spectrometer employing TMS as internal standard.

5-Chlorosulphonyl-2,4-dichlorobenzoic acid (1); was prepared according to the procedure described earlier [13].

General procedure for reaction of sulphonyl chloride (1) with nucleophiles. Preparation of sulphonamides (2-11)

To amino acid (0.1 mole), in water (25 ml) and THF (15 ml), was added triethylamine (5 ml), followed by sulphonyl chloride (1) (0.11 mole) portionwise during 30 min. The temperature of the reaction mixture during the process of addition was kept at 10°C and stirring continued 2 hrs at 20°C. Tetrahydrofuran was removed by concentration of the reaction mixture under reduced pressure, water (30 ml) added and the mixture was acidified with 2N-HCl until Conge - red (pH 5). The crude products were filtered and recrystallized from methanol - water. All the products (2-11) were chromatographically homogenous when detected with iodine and benzidine (cf. Table 1 compd. (2-11)

Some typical spectral data are as follows:

Compounds (3), (4)

IR: 3400, 3120 (NH), 1720, 1690 (C=O), 1420, 1360 (SO_2NH), 1340, 1160 (SO_2) cm^{-1}.

Compound (3)

PMR: (DMSO-d_6) δ : 12.3 (1H, COOH); 8.8 - 7.8 (2H, ArH); 5.6 (1H, NH), 1.3 - 1.1 (3H, CH_3) and other protons in support of their assigned structures.

General procedure for the synthesis of 2,4-dichlorobenzoic acid-5-sulphonylamino acid methyl ester (12-18)

A suspension of sulphonamides (2-11) (0.01 mole) in absolute methanol (150 ml) was cooled to -10°C and pure thionyl chloride (2.2 ml) was added dropwise during one hour. The reaction mixture was stirred for additional 3-4 hrs at room temperature, kept overnight and the solvent was removed in vacuum, and the residual solid material was recrystallized from methanol-water. The isolatied methyl esters (12-18) were chromatographically homogeneous when developed with benzidine (cf. Table 1 compd. (12-18). Some spectral data are as follows:

Compounds (14-15)

IR: 1760, 1725 (C=O), 1750, 1460 (-$COOCH_3$) cm^{-1}.

Compound (15)

PMR: (DMSO-d_6) δ : 3.94 (3H, $COOCH_3$) and other signals in support of their assigned structures.

General procedure for the synthesis of 2,4-dichlorobenzoic-5-sulphonylamino acid hydrazides (19-22)

The methyl ester (12-18) (0.01 mole) were dissolved in ethanol (50 ml) and hydrazine hydrate (85%) (0.05 mole) added, the reaction mixture was stirred for 3 hrs at 20°C and left to stand for 24 hrs at room temperature. The crystalline products (19-22) were filtered, washed with water and recrystallized from ethanol. The hydrazides (19-22) were chromatographically homogeneous when developed with iodine and benzidine (cf. Table 1, compd. 19-22) Some spectral data are discussed:

Compounds (19), (20)

IR: 3300, 3220 1675, 1500 ($CONH.NH_2$) cm^{-1}

Compound (20)

PMR: (DMSO-d_6) δ : 5.89 (1H, NH); 5.68 (2H, NH_2) and other protons supporting the structure of hydrazides (19-22)

General procedure for the synthesis of 2,4-dichlorobenzoylamino acid-5-sulphonyl dipeptide methyl esters (23-36)

To a solution of amino acid methyl ester hydrochloride (0.016 mole) in THF (100 ml) was added triethylamine (2 ml), the solution was stirred at 20°C for 30 min., and cooled to 0°C. 2,4-Dichlorobenzoic acid-5-sulphonylamino acid (0.008 mole) in THF (50 ml), and dicyclohexyl carbodiimide DDC (1.62 g) were added to the above mixture. The reaction mixture was stirred for 2 hrs at 0°C and for another 2 hrs at room temperature. Dicyclohexyl urea was filtered off, acetic acid (1 ml) added, the solution was left overnight and filtered, the filtrate was evaporated in vacuum. The residual material was recrystallized from methanol-water. The products (23-36) were chromatographically homogeneous when detected with benzidine.

Some spectral data are discussed:

Compounds (25),(29)

IR: 3360, 3080 (NH, CONH, SO_2NH); 1760, 1720 (C=O), and other bands due to dipeptide moieties.

Compound (25)

PMR: (DMSO-d_6) δ: 3.72 (3H, $COOCH_3$); 5.81 (H, NH).

References

1. A.M. El-Naggar, A.F. El-Haddad, S.A. El-Ghaffar, R.A. El- Sayed, *Farmaco*, 38, 7, (1985)
2. A.M. El-Naggar, F.A. Kora, R.A. El-Sayed, *J.Serb. Chem.Soc.*, 51, 441 (1986)
3. M.R. Zaher, F.A. Kora, M.E. Hussein, R.A. El-Sayed, A.M. El-Naggar, *Il-Farmaco* 41, 9 (1986).
4. F.A. Kora, M.E. Hussein, R.A. El-Sayed, A.M. Naggar, *Polish J.Chem.*, 62, 749 (1988).
5. A.M. El-Naggar, A.A. El-Saharty, *Polish J.Chem.*, 52, 2223 (1978).
6. A.M. El-Naggar, A.M. Abd-El-Salam, F.S.M. Ahmed, T.M. Ibrahim, *Acta.PharmJugosl.* 35, 15 (1985).
7. M.R. Zaher, I.M. Ismail, M.H. El-Hakim, A.M. El-Naggar, *J.Serb.Chem.Soc.*, 50, 517 (1985).
8. L. Winstein, "Sulphonamides in the pharmacological basic of therpeutics" Londong, Page 1177, (1970).
9. R.J. Cremlyn, *Int.J.Sulphur Chem.*, 8(1), 133 (1973).
10. R.J. Cremlyn, *J.Chem.Soc.(C)*, 1132 (1965).
11. R.J. Cremlyn, *J.Chem.Soc.(C)* 77, (1966)
12. R.J. Cremlyn, *J.Chem.Soc.(C)* 1341 (1969).
13. G.B. Jackman, V. Petrow, O. Stephenson and A.M. Wild, *J.Pharm.Pharmacol.* 14, 679 (1962)
14. L.J. Bellamy, 1966. "The Infrared Spectra of Complex molecules" 2nd. ed., London (1966).
15. J.G. Carlson, H.G. Dougles and H.D. Bissel, *J.Bacterial*, 55, 607.
16. G.W. Irving, T.D. Fontaine and S.P. Doolitte, *J.Bacterial* 52, 10 (1946).
17. J.V. Vincent and H.W. Vincent, *Proc. Soc. Exptl. Biol. Med.* 55, 162 (1944).