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Synthesis and Identification of Some New Tetrazole Derivatives from 4,5-dichloro Imidazole and Study of Their Biological Activity

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Abstract: A number of tetrazoles derivatives were prepared by series of steps, the first step includes react (4,5-dichloroimidazole) with (4-aminoacetophenone) to form 1-(4-((4,5-dichloro-1H-imidazol-2yl)diazenyl)ethanone (1). The second step was (1) reaction with (Amines derivatives) to get Schiff bases (2-7). The third step was reaction of Shciff base with sodiumazide to form tetrazoles derivatives (8-13). then study the biological activity for all compounds to word two type of bacteria.

Key words: Azo, Schiff base, tetrazole, imidazole, biological activity.

1. Introduction

Azo compounds are very well known and constitute one of the most varied groups of synthetic organic dyes, which are used in the textile, coloring agents for foods paper, cosmetics industries and technologies like liquid crystals [1, 2].

Azo imine compounds are important in applications different fields of science chemically and biologically. They are used in dyeing textiles, non-linear optical properties, optical switching and preparation of photoactive materials [3-5].

Tetrazole are aromatic five membered ring containing four nitrogen atoms. The first tetrazole was reported over a century ago [6-8].

Tetrazole and its derivatives have attracted much attention because of their applications as antimicrobial agents, anticancer, antifungal, a series of triazole containing tetrazole evaluated for their activity as antinociceptive and anti-inflammatory agents, anticonvulsant activity, antidiabetic activity, antihypertensive activity.

Tetrazole derivatives have considerable importance [9], its applications as antimicrobial agents, antifungal, anticancer its activity as antinociceptive [10] and anti-inflammatory [11] agents, antidiabetic activity [12], anticonvulsant activity [13, 14], antihypertensive activity [15].

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2. Materials

(FTIR) Spectra (400-4000 cm^{-1}) in KBr disk were recorded on a SHIMADZU FTIR-8400S Fourier transform. Melting points were measured using Stuart, UK. ^{13}C-NMR and ^{1}HNMR were recorded on Fourier transformation Bruker spectrometer operating at (400MHz) with (DMSO-ds) measurements were made at Department of Chemistry, Kashan University, Iran.

2.1 Synthesis Azo Derivative

1-(4-((4,5-dichloro-1H-imidazol-2-yl) diazenyl) phenyl) ethanone (1) (4-aminoacetophenone) (0.005 mmol, 0.0675 mgm) of the aromatic amine was dissolved in 5 mL of concentrated HCl and 7 mL of distilled water. The mixture is cooled to 0 °C and (0.005 mmol, 0.345) of sodiumnitrite to add dropwise with continuous stirring. The solution was left for 15 mins to stable after completing the addition (0.005

mmol) of (4,5-dichloroimidazole) dissolved in (1 mL NaOH IN 40 mL H_2O) was added, a orange precipitate was formed, filtered and recrystallization from ethanol [16].

2.2 General Method of Synthesis Schiff Base (2,3,4,5,6,7)

A mixture of equimolar quantities (0.01 mol) of (1) and (4-chloroaniline, 4-bromoaniline, 5-nitro-2-aminothiazol, 3-aminoacetophenone, 2-aminobenzimidazole, 4,4-methylenedianiline) (0.01 mol) were refluxed for 2-3 h in 5 mL of ethanol alcohol, then it was added (2) drops of glacial acetic acid as catalyst. The mixture was cooled and kept for 24 h, the crystals found were filtered, dried and recrystallization from ethanol alcohol to give (2,3,4,5,6) consecutive [17].

2.3 Synthesis of Tetrazole Derivative (8,9,10,11,13,13)

A mixture of Schiff base (2, 3, 4, 5, 6, 7) (0.001 mol) dissolved in 1,4-dioxane (15 mL) and sodium azide (0.001 mol) was dissolved in 1,4-dioxan (15 mL) and refluxed for 14-24 h. The reaction was cooled and the resulting final (8, 9, 10, 11, 12, 13) consecutive recrystallization from ethanol [18].

2.4 Preparation of Microbiology Culture Media

20 g of nutrient agar is dissolved in (500 mL) of distillation water, then put in autoclave for 20 mins at 200 °C for sterilization. Pouring the media after becoming at 37 °C in Petri dishes, made ready for streaking by bacteria. It was getting (*Escherichia coli*) and (*staphylococcus aurous*) isolated bacteria from hospital. It was cultured and these plates were incubated at 37 °C for 24 h for both bacteria [19].

3. Results and Discussion

3.1 Compound (1): 1-(4-((4,5-dichloro-1H-imidazol-2-yl)diazenyl)phenyl)ethanone

The infrared spectrum data of compound (1) showed band at (1,720) cm^{-1} for (C=O), 3,039 cm^{-1} for (Ar-H), 3,201 cm^{-1} for(N-H) imidazole, 1,666 cm^{-1} for (C=N) inside imidazole ring, 1,965 cm^{-1} for (C-H) for (CH_3), 1,480 cm^{-1} for (N=N) and 880 cm^{-1} for (C-Cl).

The ^{1}H NMR (DMSO) spectrum data of compound (1) show δ: 6.9-8.1 (m.,4H, Ar-H),4.2 (S, 3H, CH_3), 11.2 (S, 1H, N-H imidazole ring).

The ^{13}C-NMR (DMSO) spectrum data of compound (1) show δ: 149 (C3,C_2), 197 (C_{10}), 157(C_1), 26 (C_{11}), 135(C_7), 133(C_4), 130-110 C arom.

3.2 Compound (2): 4-chloro-N-(1-(4-((4,5-dichloro-1H-imidazol-2-yl)diazenyl)phenyl)ethylidene)aniline

The infrared spectrum data of compound (2) showed band at 3,055 cm^{-1} for (Ar-H), 3,116 cm^{-1} for (N-H) imidazole, 1,610 cm^{-1} for (C=N), 1,458 cm^{-1} for (N=N), 650 cm^{-1} for (C-Br).

3.3 Compound (3): 4-bromo-N-(1-(4-((4,5-dichloro-1H-imidazol-2yl)diazenyl)phenyl)ethylidene)aniline

The infrared spectrum data of compound (3) showed band at 3,055 cm^{-1} for (Ar-H), 3,116 cm^{-1} for (N-H) imidazole, 1,610 cm^{-1} for (C=N), 1,458 cm^{-1} for (N=N) 650 cm^{-1} for (C-Br). The ^{1}H NMR (DMSO) spectrum data of compound (3) show δ: 7.1-7.8 (m., 8, Ar-H), 10.7 (S, 2H, NH imidazol ring), 1.9 (S, 3H, CH_3).

The ^{13}C-NMR (DMSO) spectrum data of compound (3) show δ: 141 (C_3, C_2), 163 (C_{10}), 157 (C_{12}), 149 (C_1), 61 (C_{11}), 1,268 (C_{13}), 132 (C_7), 130 (C_4), 120-142 Carom.

3.4 Compound (4): N-(1-(4-((4,5-dichloro-1H-imidazol-2-yl)diazenyl)phenyl)ethylidene)-5-nitrothiazol-2-amine

The infrared spectrum data of compound (4) showed band at 1,250 cm^{-1} for (C-S), 3,016 cm^{-1} for (Ar-H), 2,180 cm^{-1} for (N-H) imidazole, 1,596 cm^{-1} for (C=N) inside imidazole ring, 836 cm^{-1} for (C-Cl) 1,566 cm^{-1} for (C=C) Aromatic.

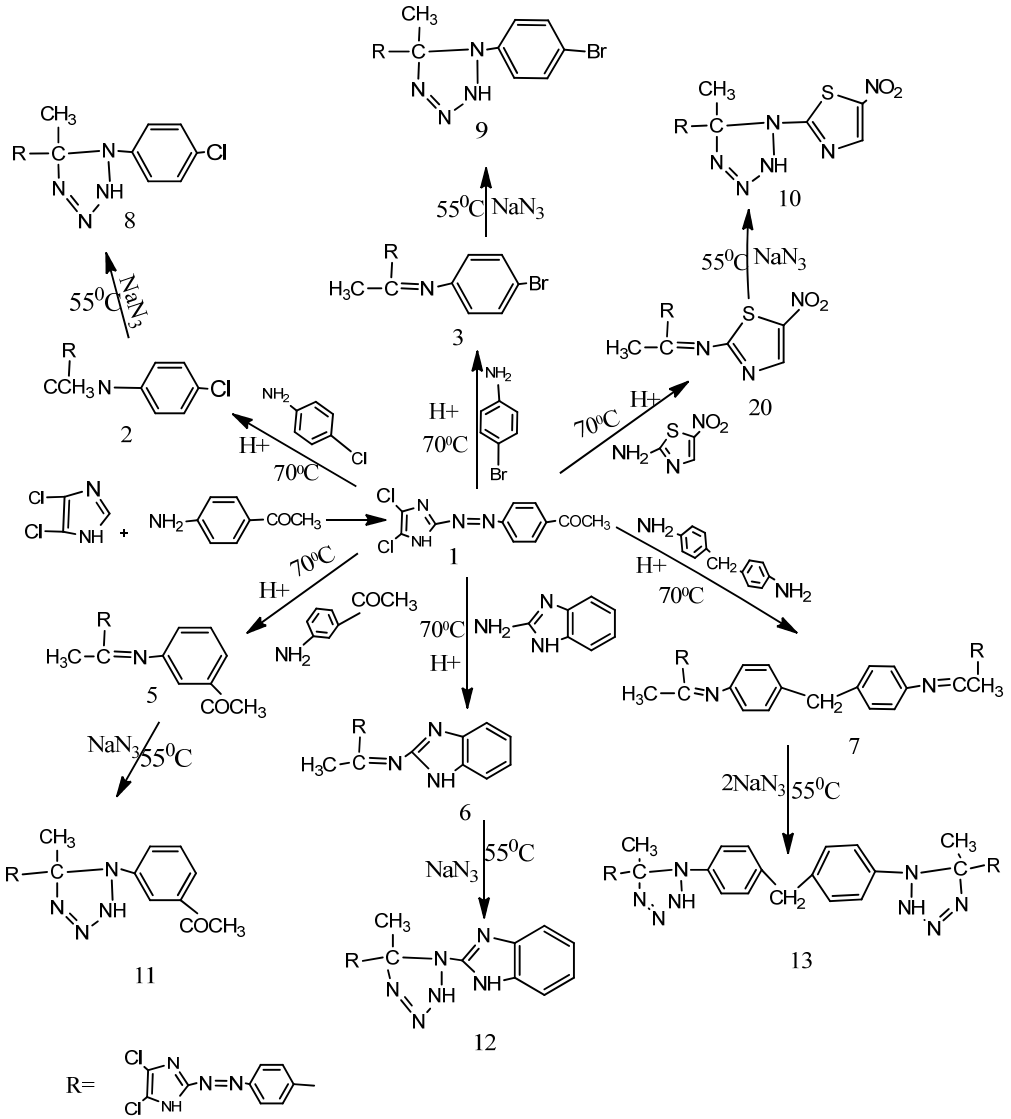

Fig. 1 Scheme of preparation Tetrazole derivatives.

3.5 Compound (5): 1-(3-((1-(4-((4,5-dichloro-1H-imidazol-2-yl)diazenyl)phenyl)ethylidene)amino)phenyl) ethanone

The infrared spectrum data of compound (5) showed band at 1,740 cm^{-1} for (C=O), 3,040 cm^{-1} for (Ar-H), 3,220 cm^{-1} for (N-H) imidazole, 1,580 cm^{-1} for (C=N) inside imidazole ring, 2,955 cm^{-1} for (H-H) in CH_3 1,566 cm^{-1} for (C=C) Aromatic.

3.6 Compound (6): N-(1-(4-((4,5-dichloro-1H-imidazol-2-yl)diazenyl) phenyl) ethylidene)-1H-benzo [d] imidazol-2-amine

The infrared spectrum data of compound (6) showed band at 3,020 cm^{-1} for (Ar-H), 3,160 cm^{-1} for (N-H) imidazole, 1,566 cm^{-1} for (C=N) inside imidazole ring, 1,546) cm^{-1} for (C=C)Aromatic.

3.7 Compound (7): 4,4'-methylenebis(N-(1-(4-((4,5-dichloro-1H-imidazol-2-yl)diazenyl)phenyl)ethylidene) aniline)

The infrared spectrum data of compound (7) showed band at 2,931 cm^{-1} for (C-H) in (CH_3), 3,024 cm^{-1} for (Ar-H), 3,132 cm^{-1} for(N-H) imidazole, 1,598 cm^{-1} for (C=N) inside Imidazole ring, 840 cm^{-1} for (C-Cl), 1,566 cm^{-1} for (C=C) Aromatic.

3.8 Compound (8): 2-(1H-benzo [d] imidazol-2-yl) -4-(5-(4-(dimethylamino)phenyl)-1H-tetrazol-1-yl)phenol

The infrared spectrum data of compound (8) showed band at 3,178 cm^{-1} for (N-H) in tetrazole, 3,055 cm^{-1} for (Ar-H), 2,977 cm^{-1} for (C-H) in CH_3, 1,566 cm^{-1} for (C=C) Aromatic.

The ^{1}H NMR (DMSO) spectrum data of compound (8) show δ: 6.5-7.6 (m., 8H, Ar-H), 10.3 (S, 1H, H-N Tetrazole) 10.1 (S, 1H, N-H) imidazole.

The ^{13}C-NMR (DMSO) spectrum data of compound (8) show δ: 125 (C_3, C_2), 137 (C_{12}), 128 (C_{15}), 130 (C_1), 66 (C_{11}), 129 (C_7), 126 (C_3, C_4).

3.9 Compound (9): 1-(4-bromophenyl)-5-(4-((4,5-dichloro-1H-imidazol-2-yl)diazenyl)phenyl)-5-methyl-2,5-dihydro-1H-tetrazole

The infrared spectrum data of compound (9) showed band at 3,180 cm^{-1} for (N-H) intetrazole, 3,016 cm^{-1} for (Ar-H), 2,977 cm^{-1} for (C-H) in CH_3, 1,419 cm^{-1} for (N=N), 1,566 cm^{-1} for (C=C) Aromatic.

The ^{1}H NMR (DMSO) spectrum data of compound (9) show δ: 6.5-7.8 (m., 8H, Ar-H), 9.6 (S, 1H, NH Amidazol ring), 10.5 (S, 1HN-H tetrazole, 3.8 (S, 3H, CH_3).

The ^{13}C-NMR (DMSO) spectrum data of compound (9) show δ: 158 (C_{10}), 155 (C_{12}), 160 (C_1), 62 (C_{11}), 137 (C_1), 123 (C_6, C_7), 148 (C_4) 133-112 (Carom).

3.10 Compound (10): 2-(5-(4-((4,5-dichloro-1H-imidazol-2-yl)diazenyl)phenyl)-5-methyl-2,5-dihydro-1H-tetrazol-1-yl)-5-nitrothiazole

The infrared spectrum data of compound (10) showed band at 3,223 cm^{-1} for (N-H) in tetrazole, 3,055 cm^{-1} for (Ar-H), 1,450 cm^{-1} for (N=N) inside imidazole ring, 840 cm^{-1} for (C-Cl).

The ^{1}H NMR (DMSO) spectrum data of compound (10) show δ: 7-8.3 (m., 5H, Ar- H), 1.2 (S, 3H, CH_3), 9.1 (S, 1H, NH Amidazol ring), 9.3 (1H, NH, tetrazole).

The ^{13}C-NMR (DMSO) spectrum data of compound (10) show δ: 147 (C_{12}), 125.2 (C_1), 128.7 (C_{11}), 128.7 (C_{13}) 128.14 (C_{14}), 112-125 (Carom).

3.11 Compound (11): 1-(3-(5-(4-((4,5-dichloro-1H-imidazol-2-yl)diazenyl)phenyl)-5-methyl-2,5-dihydro-1H-tetrazol-1-yl)phenyl)ethanone

The infrared spectrum data of compound (11) showed band at 3,224 cm^{-1} for (N-H) in tetrazole, 3,016 cm^{-1} for (Ar-H), 3,163 cm^{-1} for (N-H) imidazole, 1,419 cm^{-1} for (N=N), 636 cm^{-1} for (C-Br).

The ^{1}H NMR (DMSO) spectrum data of compound (11) show δ: 6.2-8 (m., 8H, Ar- H), 10.1 (S, 1 H, N-H tetrazole), 9.8 (S, 1H, NH Amidazol ring), 3.2 (S, 3H, OCH_3), 1.3 (S, 3H, CH_3).

The ^{13}C-NMR (DMSO) spectrum data of compound (11) show δ: 113 (C_5, C_2), 137 (C_{10}), 157 (C_{12}), 190 (C_{18}), 138 (C_1), 61 (C_{11}), 62 (C_{19}), 141 (C_7).

3.12 Compound (12): 42-(5-(4-((4,5-dichloro-1H-imidazol-2-yl)diazenyl)phenyl)-5-methyl-2,5-dihydro-1H-tetrazol-1-yl)-1H-benzo[d]imidazole

The infrared spectrum data of compound (12) showed band at 3,130 cm^{-1} for (N-H) in tetrazole, 3,016 cm^{-1} for (Ar-H), 1,450 cm^{-1} for (N=N), (1,350, 1,500) cm^{-1} for (NO_2), 1.2 (s, 3H, CH_3).

The ^{1}H NMR (DMSO) spectrum data of compound (12) show δ: 6-7.8 (m., 6H, Ar-H), 10.4 S, 1 H OH, 5.4 (S, 2H, NH_2), 10.6 (S, 2H, NH Amidazol ring).

The ^{13}C-NMR (DMSO) spectrum data of compound (12) show δ: 158 (C_{12}), 128 (C_8), 160 (C_1), 66 (C_{11}), 130 (C_1), 129 (C_7), 129 (C_{18}, C_{17}), 111-125 (Carom).

3.13 Compound (13): bis(4-(5-(4-((4,5-dichloro-1H-imidazol-2-yl)diazenyl)phenyl)-5-methyl-2,5-dihydro-1H-tetrazol-1-yl)phenyl)methane

The infrared spectrum data of compound (13) showed band at 3,200 cm^{-1} for (N-H) in tetrazole, 3,055 cm^{-1} for (Ar-H), 1,411 cm^{-1} for (N=N), 2,923 cm^{-1} for (C-H) in CH_3.

The ^{1}H NMR (DMSO) spectrum data of compound (13) show δ: 6.2-8 (m, 16H, Ar- H), 10.1 (S, 1 H, N-H, tetrazole), 3.4 (S, 2H, CH_2), 9.2 (S, 1H, NH Amidazol

ring), 1.2 (S, 6H, CH_3).

The ^{13}C-NMR (DMSO) spectrum data of compound (13) show δ: 145 (C_{25}, C_{10}), 61 (C_{26}, C_{11}), 66 (C_{18}), 163 (C_{12}, C_{22}), 159 (C_1, C_{33}), 111-141 (Carom).

4. Conclusions

In this work, the antibacterial test was performed according to the wells method. Compounds (1-30) were assayed for their antimicrobial activity in vitro against Gram-negative bacteria (*Escherichia coli*) and Gram-positive bacteria *(staphylococcus aurous)*. Prepared agar and Petri dishes were sterilized by autoclaving for 20 min at 200 °C. These plates were incubated at 37 °C for 24 h for both bacteria. DMSO was used as a solvent to prepare solutions of the various compounds were examined (0.02 g of comp./5 mL DMSO). The inhibition zones caused by the various compounds were examined, where compounds which appeared good activity (1, 8, 18 and 28) against *(staphylococcus aurous*) on other hand; compounds (1 and 13) which appeared good activity against (*Escherichia coli*). The results of the preliminary screening tests are listed in Table 1 [25].

Table 1 Biological activity for compounds (1-13).

Compounds No.	*E. coli*	*Staph. aureus*	Compounds No.	*E. coli*	*Staph. aureus*
1	+	+	8	R	+++
2	+	+++	9	R	+++
3	-	++	10	R	-
4	-	++	11	R	++
5	-	-	12	R	+++
6	-	-	13	+	++
7	-	+++			

- = No inhibition = inactive , + = (5-10) mm = slightly active , Resistance(+-) = R

++ = (11-20) mm = moderately active, +++ = (more than 20) mm = Good active.

Table 2 Physical properties of compounds (1-13).

No.	Name of comp.	M. F	M. W	M.P (°C)	R_f	Colour	%
1	1-(4-((4,5-dichloro-1H-imidazol-2-yl) diazenyl) phenyl)ethanone	$C_{11}H_8Cl_2N_4O$	283	218-220	0.3	Orange	83
2	4-chloro-N-(1-(4-((4,5-dichloro-1H-imidazol-2-yl) diazenyl)phenyl)ethylidene)aniline	$C_{17}H_{12}Cl_3N_5$	392	160-158	0.3	Black	76
3	4-bromo-N-(1-(4-((4,5-dichloro-1H-imidazol-2-yl) diazenyl)phenyl)ethylidene)aniline	$C_{17}H_{12}BrCl_2N_5$	437	150-148	0.2	Black	88
4	N-(1-(4-((4,5-dichloro-1H-imidazol-2-yl) diazenyl)phenyl)ethylidene)-5-nitrothiazol-2-amine	$C_{14}H_9Cl_2N_7O_2S$	410	186-184	0.2	Brown	80
5	1-(3-((1-(4-((4,5-dichloro-1H-imidazol-2-yl) diazenyl)phenyl)ethylidene)amino)phenyl)ethanone	$C_{19}H_{15}Cl_2N_5O$	400	145-143	0.4	Violt	71
6	N-(1-(4-((4,5-dichloro-1H-imidazol-2-yl)diazenyl)phenyl)ethylidene) -1H-benzo[d]imidazol-2-amine	$C_{18}H_{13}Cl_2N_7$	398	175-173	0.3	Red	78
7	4,4'-methylenebis(N-(1-(4-((4,5-dichloro-1H-imidazol-2-yl) diazenyl)phenyl)ethylidene)aniline)	$C_{35}H_{26}Cl_4N_{10}$	728	174-175	0.4	Black	71
8	1-(4-chlorophenyl)-5-(4-((4,5-dichloro-1H-imidazol-2-yl) iazenyl)phenyl)-5-methyl-2,5-dihydro-1H-tetrazole	$C_{17}H_{13}Cl_3N_8$	434	150-148	0.4	Brown	77
9	1-(4-bromophenyl)-5-(4-((4,5-dichloro-imidazol-2-yl) diazenyl)phenyl)-5-methyl-2,5-dihydro-1H-tetrazole	$C_{17}H_{13}BrCl_2N_8$	480	180-178	0.3	Brown	76
10	2-(5-(4-((4,5-dichloro-imidazol-2-yl)diazenyl)phenyl) -5-methyl-2,5-dihydro-1H-tetrazol-1-yl)-5-nitrothiazole	$C_{14}H_{10}Cl_2N_{10}O_2S$	453	300-298	0.4	Brown	80
11	1-(3-(5-(4-((4,5-dichloro-imidazol-2-yl) diazenyl) phenyl) -5-methyl-2,5-dihydro-1H-tetrazol-1-yl)phenyl) ethanone	$C_{19}H_{16}Cl_2N_8O$	443	150-148	0.5	Brown	68
12	2-(5-(4-((4,5-dichloro-imidazol-2-yl)diazenyl)phenyl) -5-methyl-2,5-dihydro-1H-tetrazol-1-yl)-1H-benzo[d]imidazole	$C_{18}H_{14}Cl_2N_{10}$	441	157-155	0.4	Black	72
13	bis(4-(5-(4-((4,5- -1H-imidazol-2-yl)diazenyl)phenyl) -5-methyl-2,5-dihydro-1H-tetrazol-1-yl)phenyl)methane	$C_{35}H_{30}Cl_2N_{16}$	814	165-163	0.4	Red	71

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