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Synthesis, Characterization And Evaluation of Antimicrobial of Some new Azo compounds derived from 2-amino-6-methoxybenzothiazole

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Abstract:

This research include the preparation of Schiff bases by the reaction of (2-amino-6-methoxybenzothiazole) with *p*-aminoacetophenone to product compound [A], and then converted to Azo compounds from the reaction of Schiff base [A] with *p*-vanillin to product compound [B],and then reacted with different aromatic amines . These compounds character FT-IR ,^{1}HNMR,^{13}C-NMR and Melting points and evaluate their Antimicrobial activity on four types of bacteria , (*S. aureus and S. epidermidis*) (*K. pneumonia and E. coli*) .

Keywords: *Azo compounds, 2-amino-6-methoxybenzothiazole, Antimicrobial*

1.Introduction

The compensated benzothiazole ring has a distinctive pharmacological importance and structure in medical chemistry, Thiazole is one of the active chemicals in both medicine and biology since it is made up of the fusion of the benzene and thiazole rings, as well as the sulfur and nitrogen atoms that make up its basic structure[1]. Replacement benzothiazole rings play a significant role in the chemical structure of medications used to treat a variety of illnesses[2], and they have beneficial therapeutic effects such as anti-tuberculosis[3], anti-malaria[4], antimicrobial[5], antitumor[6], anthelmintic[7], analgesic[8], anti-inflammatory[9], and antidiabetic properties[10], Compounds containing the isomethin group (-CH=N-) are known as Schiff bases, and are prepared as a result of the condensation of primary aromatic amine with aldehyde or ketone [11]and Schiff bases are known by different names such as Azomethin, Benzanils, and imins. Azo compounds were discovered by the scientist (Perkin) in 1856, who succeeded in preparing the synthetic azo dye and called it (mauveine) Moven[12], azo compounds contain the active group (- N = N-) and called the azo group is of commercial importance because it constitutes more than half of the commercial dyes, giving permanent colors when placed on fibers, Azo dyes were frequently used to color paper, textile fibers, food, and cosmetics., It is characterized by its ease of preparation and distinctive colors[13].

ISSN: 0975-3583,0976-2833 VOL14, ISSUE 08, 2023

2.Experimental

2.1.Materials and devices used

All solvents and reagents were used through this work purchased from Aldrich, CDH Companies. Melting points are uncorrected and were recorded in an open capillary tube on Stuart melting point apparatus. Infrared spectra have been recorded on a Shimadzo FTIR-8100 spectra using KBr discs– and ^{1}H-NMR Spectra have been measured on a MHz spectra using (DMSO-d^{6}) as solvent. reaction monitoring and verification of the purity of the compounds was done by TLC on silica gel-percolated alumni sheets (type 60 F254 Merck, Darmstadt, Germany).

2.2.Synthesis of compounds [A][14]

(3.6g, 0.02mol) of 2-amino-6-methoxybenzothiazole was mixed in (15ml) of absolute ethanol and (4) drops of ice acetic acid were added to it in a circular flask with a capacity of 50 ml and then added to it (2.7g, 0.02mol) of p-aminoacetophenone dissolved in absolute ethanol with continuous stirring and escalation for (6) hours and it was confirmed that the reaction ended using TLC technology and after the end of the reaction the resulting solution was poured into an hour bottle and left to dispose of the solvent, The precipitate was collected, dried and recrystallized using ethanol. The molecular formula of the product $C_{16}H_{15}N_3OS$ was 285.37 mol/g with a light brown color and melting point (95-97) ^{o}C and a percentage of 75%, and the handicap coefficient was 0.61 (R_f).

2.3.Synthesis of azo compounds [B][15]

Dissolve (2.97g, 0.01mol) of the compound [A] in a conical flask in (30ml) of 37% HCl acidic solution at a temperature of (0-5^{o}C) with continuous stirring. Dissolved in another flask (1.52g, 0.02mol) of $NaNO_2$ in a small amount of distilled water (3ml) and then added to the first flask solution at a temperature of (0-5^{o}C) with continuous stirring and the addition is gradual in the form of drops as it was observed color change evidence of the formation of diazonium salt. The coupling solution was prepared where Dissolve (2.73g, 0.01mol) of p-vanillin in (10ml) of pyridine at a temperature of (0-5^{o}C) and after the completion of the dissolution through continuous stirring, add the diazonium salt prepared in the first step with stirring for a period of - an hour, The reaction mixture was poured into 100 ml of cold water, then the mixture was filtered, washed with cold water and then recrystallized using ethanol, and its molecular formula was $C_{24}H_{20}N_4O_4S$, molecular weight 460.5 mol/g in dark brown color, melting point (114-112) ^{o}C and its percentage was 77%, and the disability coefficient was 0.48 (R_f).

ISSN: 0975-3583,0976-2833 VOL14, ISSUE 08, 2023

2.4.Synthesis of compounds [C1-C10][16]

Dissolve (1.38g, 0.003mol) of the compound [B] in 15ml of absolute ethanol with the addition of (4) drops of ice acetic acid and then add (3mmol) of aromatic amines to the mixture in a circular flask. Stir for 5-7 hours with constant stirring. The end of the reaction and the purity of the product using TLC technique were confirmed after the end of the reaction by cooling the resulting mixture and leaving it in a watch bottle to evaporate the alcohol and collect the precipitate. It was dried and then recrystallized using absolute ethanol, the Rf, percentage, and physical characteristics of the produced compounds [C1-C10] are presented in Table (1).

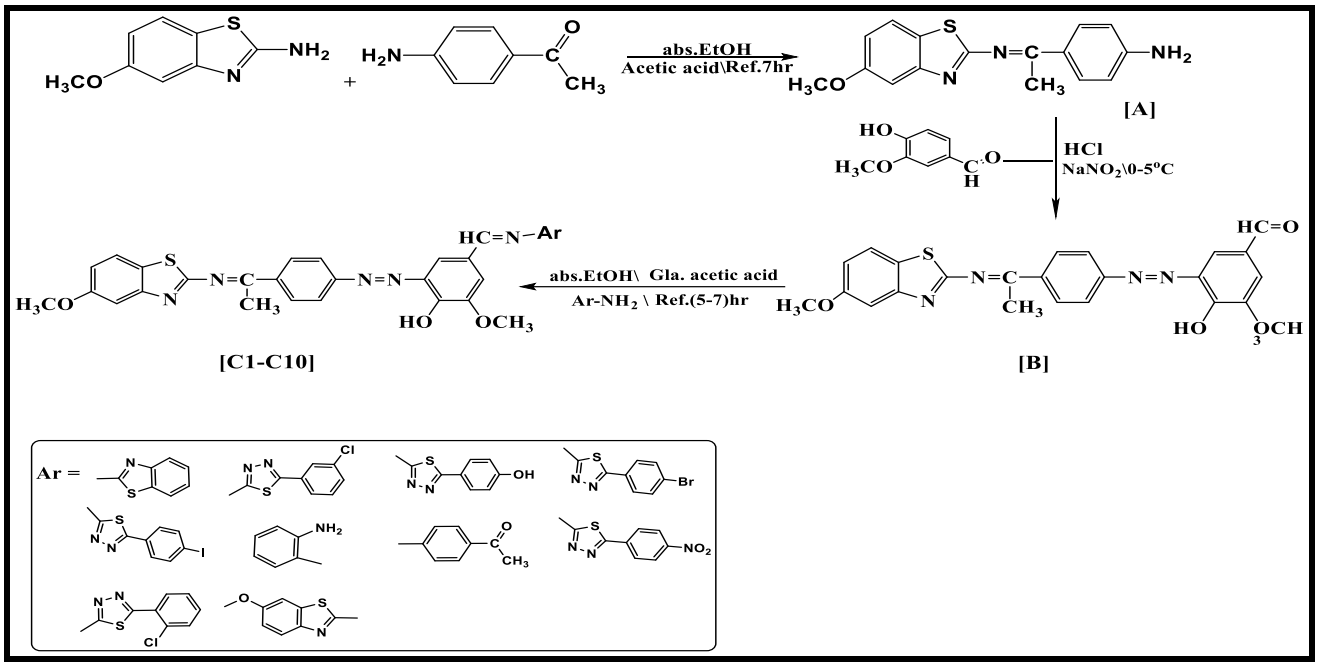
2.5. Evaluation of Antimicrobial activity of[C1-C10][17]

This medium was made by dissolving 38 grams in 1L of distillated water, sterilizing it in an autoclave at 121 °C under 15 pounds of pressure for 15 minutes, cooling it down, pouring it onto sterile plates, and refrigerating it until it was needed. Using a sterilized cork borer, holes with a diameter of 5 mm were formed in the culture medium. Individually, 100 l of the material (concentration 100, 50 mg/ml) was applied to each hole using a micropipette. After that, incubate the dishes for 24 hours at 37 °C. Nitrofurantoin (300 g) was placed in the center of Petri dishes containing Staphylococcus aureus and Staphylococcus epidermidis, whereas Nalidixic acid (30 g) was placed in the center of dishes containing E. coli and Klebsiella pneumonia..

3.Results and Discussion:

In this research prepared Schiff bases compounds [A], azo [B] and other compounds containing azo dye [C1-C10] and were characterization by FT-IR, ^{1}H-NMR and ^{13}C-NMR as in Scheme(1).

ISSN: 0975-3583,0976-2833 VOL14, ISSUE 08, 2023



Scheme(1): Synthesis of Azo compounds

3.1.Characterization of compound [A]

When studying the infrared spectrum (FT-IR)[18] of the compound [A], it was noted that two absorption beams belonging to symmetrical and asymmetric stretching appeared at the frequencies (3332,3387)cm^{-1} for the (NH_2) group respectively, and an absorption beam appeared at (3101)cm^{-1} belonging to the aromatic (C-H) bond, and also the appearance of two symmetrical and asymmetrical stretch beams for the Aliphatic (C-H) group and appeared at (2846,2905)cm^{-1} respectively, and the spectrum also showed a sharp and medium absorption band at (1651)cm^{-1} belonging to the group (C = N), which is evidence of the composition of the product. Two absorption beams belong to symmetrical and asymmetrical stretching respectively for the aromatic (C=C) at (1450,1558)cm^{-1} and the appearance of two stretch beams at (1273,1188)cm^{-1} belonging to the ether group (O-C-O) and a bundle at (1118)cm^{-1} belonging to the (C-N) group, as shown in the **Figure**(1).

3.2. Characterization of 4-hydroxy-3-methoxy-5-((4-(1-((5-methoxybenzo[d]thiazol-2-yl)imino)ethyl)phenyl)diazenyl)benzaldehyde [B]

When studying the infrared spectrum (FT-IR)[19] of the compound [B], it was noted that the two absorption beams belonging to the frequency of the stretching group (NH_2) and the appearance of a wide and high-intensity absorption beam at (3425)cm^{-1} were due to the elicitation of the (OH) group and the elastic absorption bundle of the aromatic C-H at (3070)cm^{-1} and the appearance of two symmetrical and asymmetrical stretch beams and respectively belonging to the Aliphatic group (C-

ISSN: 0975-3583,0976-2833 VOL14, ISSUE 08, 2023

H) at (2831,2939) cm^{-1} respectively and an absorption bundle at (2738)cm^{-1} due to the elicitation of the elastic (C-H) Aldehyde, and we note the appearance of a sharp and strong beam at (1689) cm^{-1} belongs to the carbonyl group in aldehyde (C=O), as well as a sharp absorption bundle appeared for the group (C=N) at (1666) cm^{-1}, and two absorption bundles belonging to the elasticity (C=C) appeared at (1512.1597) cm^{-1}, while the stretch bundle of the group (N=N) appeared at (1465) cm^{-1}, and it was noted that two symmetrical and asymmetrical stretch beams appeared at (1265.1087) cm^{-1} belonging to the (O-C-O) etheric group shown in the **Figure(2).**

3.3.Characterization of Azo compounds [C1-C10]

When studying the infrared spectrum (FT-IR)[20] of the prepared compounds [C1-C10], it was observed that the carbonyl group stretch beam (C=O) of the aldehyde that appeared at (1689) cm^{-1} disappeared, and the wide absorption beams belonging to the OH stretch at the range (3465-3317) cm^{-1}, and the elastic beam of the aromatic (C-H) of the benzene ring at the range (3001-3087)cm^{-1}, and the appearance of two asymmetrical and symmetrical stretch beams at (2916-2978)cm^{-1} and (2831-2893)cm^{-1} respectively belonging to the aliphatic C-H stretch. Also, an absorption beam of elastic stretching (C=N) appeared at the range (1654-1675) cm^{-1}, and also two absorption beams appeared at the range (1504-1550)cm^{-1} and (1550-1604) cm^{-1} belonging to the elastic stretch (C=C) aromatic, and showed an absorption bundle of elastic stretching (N=N) at the range (1412-1485)cm^{-1}, and two absorption packages belonging to the elastic stretch (C-O-C) at the range (1058-1234)cm^{-1} (1219-1273)cm^{-1} shown in the **Table(2)** and **Figure(3)**

the proton nuclear magnetic resonance spectrum (^{1}H-NMR)[21] of the compound [C5], it was noted that a signal appeared at (1.90) ppm belonging to (CH_3)protons, and two single signals appeared at (3.76,3.72)ppm to (OCH_3)group, and a binary (6.81-8.02)ppm belonging to protons Aromatic rings (Ar-CH), and a single signal also appeared at (8.95)ppm Azomethene group (HC=N), as well as (9.18)ppm to (OH) protons, shown in the **Figure(4)** of compound [C5]

the nuclear magnetic resonance spectrum of carbon (^{13}C-NMR)[22] of the compound [C5], also showed a signal at (26.37)ppm belonged to the methyl group carbon atom (CH_3), and that the two signals at the chemical displacement (55.49,57.02) ppm belong to two carbon atoms of the methoxy group (OCH3) in the benzothiazole ring and the p-vanillin ring, and one signal also appeared at (97.58)ppm belonging to the carbon atom of the (C-I) group in the benzene ring. The spectrum also showed single signals at chemical displacements range (105.36-146.78)ppm belong to atoms of aromatic rings (Ar-CH), and also the appearance of two single signals at chemical displacement (155.62,154.22)ppm belong to two atoms of the carbon group azomethene (C = N-H), and that the two signals at chemical displacements (169.71,164.71) ppm belong to the carbon atoms

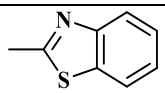
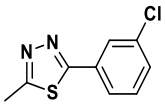
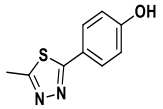
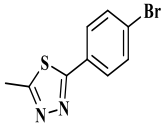
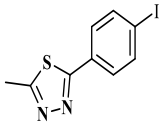
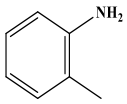
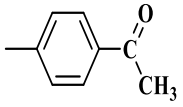
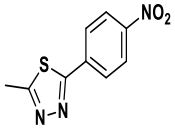
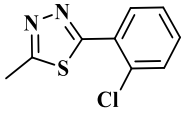
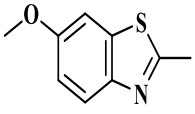
ISSN: 0975-3583,0976-2833 VOL14, ISSUE 08, 2023

group (C = N) of the benzothiazole ring and 4-3-1-benzodiazole,as shown in the **Figure(5)** of compound [C5]

3.4. Antimicrobial activity of [C1-C10]

Were Studied the Antimicrobial activity of some compounds[C1][C2][C3][C4][C8][C9] prepared against two types of bacterial isolates Gram-positive for the dye (Gr^{+ve}) (***S. aureus and S. epidermidis***) and two types Gram-negative for the dye (Gr^{-ve}) (***K. pneumonia and E. coli***) use three concentrations (100, 50, 25) mg/mL in studied Some of the prepared compounds have shown good inhibitory activity against the bacteria used[23], as shown in the **Table(3), Scheme(2,3).**

Table (1): Physical characteristic of synthesized compounds[C1-C10]

No.	*Ar*	*Formula*	*M.wt*	*Color*	*M.P(°C)*	*Yield %*	R_f
C1		$C_{31}H_{24}N_6O_3S_2$	592.6	Gray	95-97	60	0.42
C2		$C_{32}H_{24}ClN_7O_3S_2$	654.1	Light Brown	115-117	72	0.46
C3		$C_{32}H_{25}N_7O_4S_2$	635.7	Red	117-119	55	0.40
C4		$C_{32}H_{24}BrN_7O_3S_2$	698.6	Dark brown	113-115	68	0.39
C5		$C_{32}H_{24}IN_7O_3S_2$	745.6	Dark Brown	120-122	64	0.41
C6		$C_{31}H_{26}N_6OS$	530.6	Brown	112-115	70	0.52
C7		$C_{32}H_{27}N_5O_4S$	577.6	Black	105-107	57	0.53
C8		$C_{32}H_{24}N_8O_5S_2$	664.7	Dark brown	114-115	75	0.39
C9		$C_{32}H_{24}ClN_7O_3S_2$	654.1	Red	118-120	69	0.41
C10		$C_{36}H_{30}N_6O_5S_2$	690.7	Reddish brown	109-111	53	0.47

ISSN: 0975-3583,0976-2833 VOL14, ISSUE 08, 2023

Table (2): FT-IR data of synthesized compounds[C1-C10]

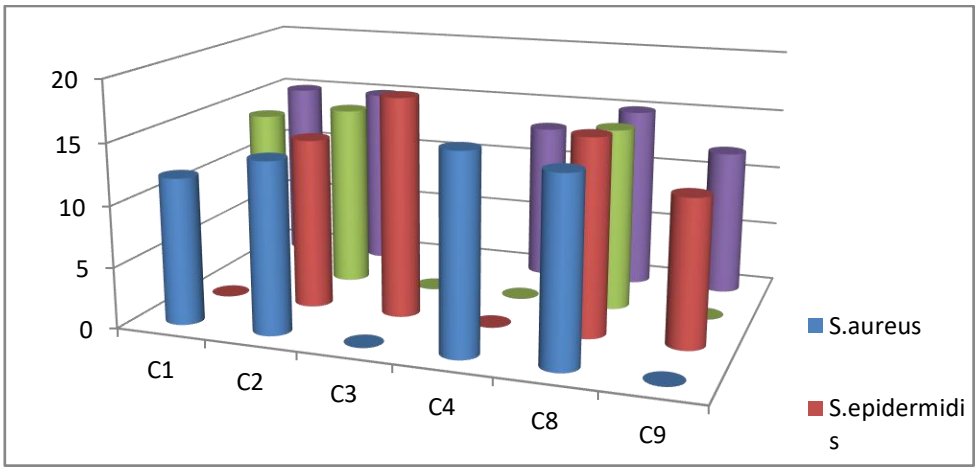
Comp. No.	OH	C-H Arom.	C-H Aliph.	C=N	N=N	C=C Arom.	C-O-C)	Others
C1	3317	3062	2831 2924	1666	1450	1535 1604	1265 1172	----
C2	3433	3008	2831 2939	1658	1473	1504 1597	1265 1234	υ C-Cl(540)
C3	3371	3047	2893 2978	1667	1481	1527 1558	1249 1167	----
C4	3309	3071	2839 2962	1666	1465	1512 1597	1265 1134	νC-Br(686)
C5	3340	3001	2831 2949	1657	1412	1543 1595	1257 1224	νC-I(550)
C6	3348	3086	2846 2916	1675	1460	1512 1604	1266 1156	NH_2 (Sym)(3240) (Asy)(3326)
C7	3464	3055	2831 2970	1673	1481	1512 1589	1273 1114	υ C=O(1743)
C8	3410	3078	2831 2939	1674	1485	1543 1604	1265 1219	υ NO_2 Asy(1519) Sym(1388)
C9	3332	3086	2862 2954	1654	1427	1519 1604	1265 1126	υ C-Cl (524)
C10	3387	3001	2839 2955	1663	1435	1504 1560	1273 1132	----

Table (3): Antibacterial efficacy of some prepared compounds (inhibition circle diameter in mm)

Comp. No.	Concentration mg/mL	Gram-positive		Gram-negative	
		S. aureus	***S. epidermidis***	***E. coli***	***K. pneumonia***
C1	100	12	0	14	15
	50	19	11	0	11
	25	0	0	0	0
C2	100	14	14	15	15
	50	12	0	12	11
	25	0	0	0	0

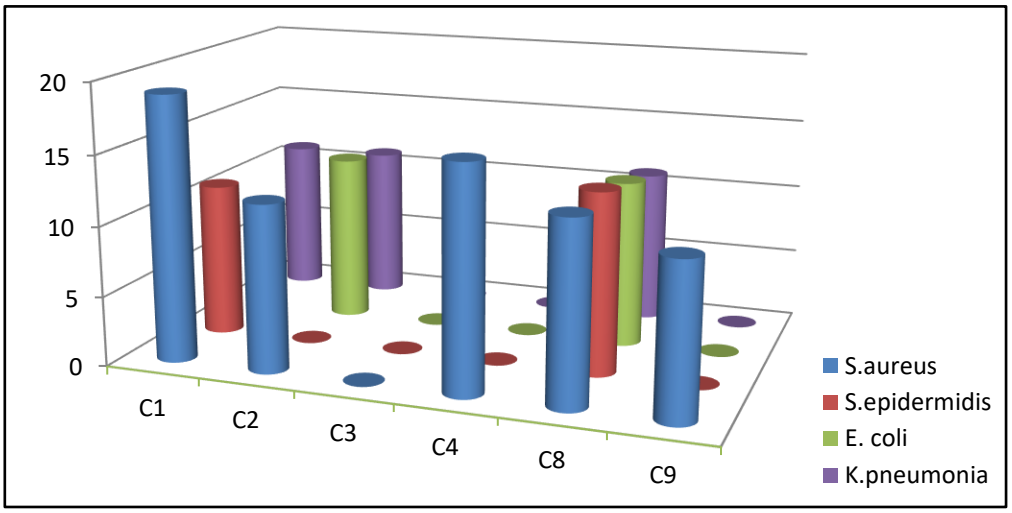
ISSN: 0975-3583,0976-2833 VOL14, ISSUE 08, 2023

	100	16	18	0	12
C3	50	0	0	0	0
	25	0	18	0	0
	100	12	0	0	13
C4	50	16	0	0	0
	25	0	0	0	0
	100	15	16	15	15
C8	50	13	13	12	11
	25	0	0	0	0
	100	0	12	0	12
C9	50	11	0	0	0
	25	0	0	0	0
Nalidixic acid	30	–	–	11	0
Nitrofurantoin	300	17	15	–	–

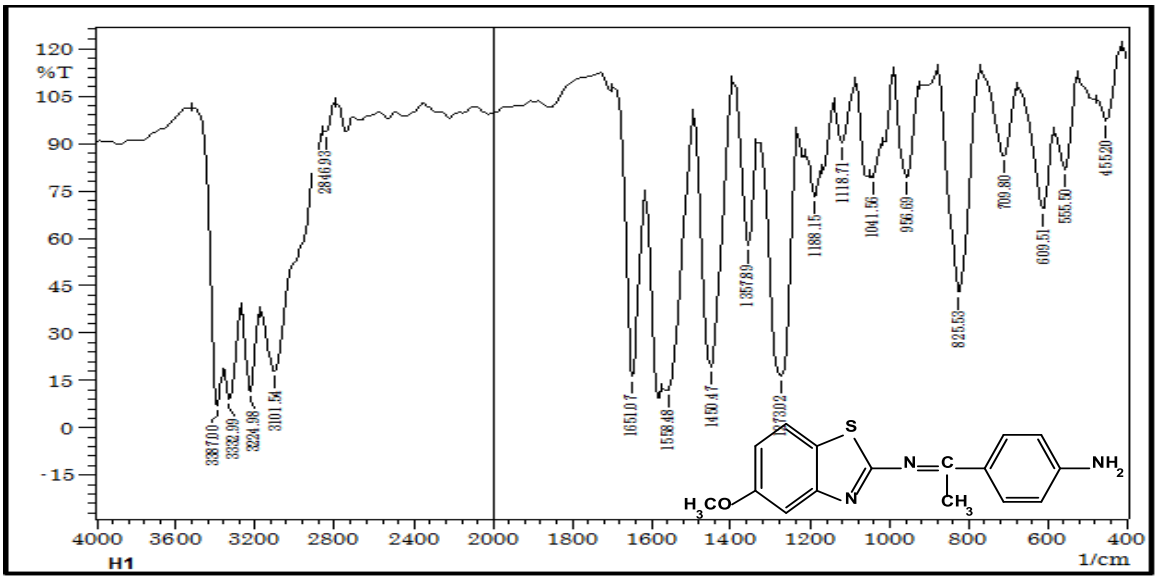


Scheme(2): Biological Activity Values of Compounds Prepared Against Four Types of Bacteria at 100 mg/mL

ISSN: 0975-3583,0976-2833 VOL14, ISSUE 08, 2023

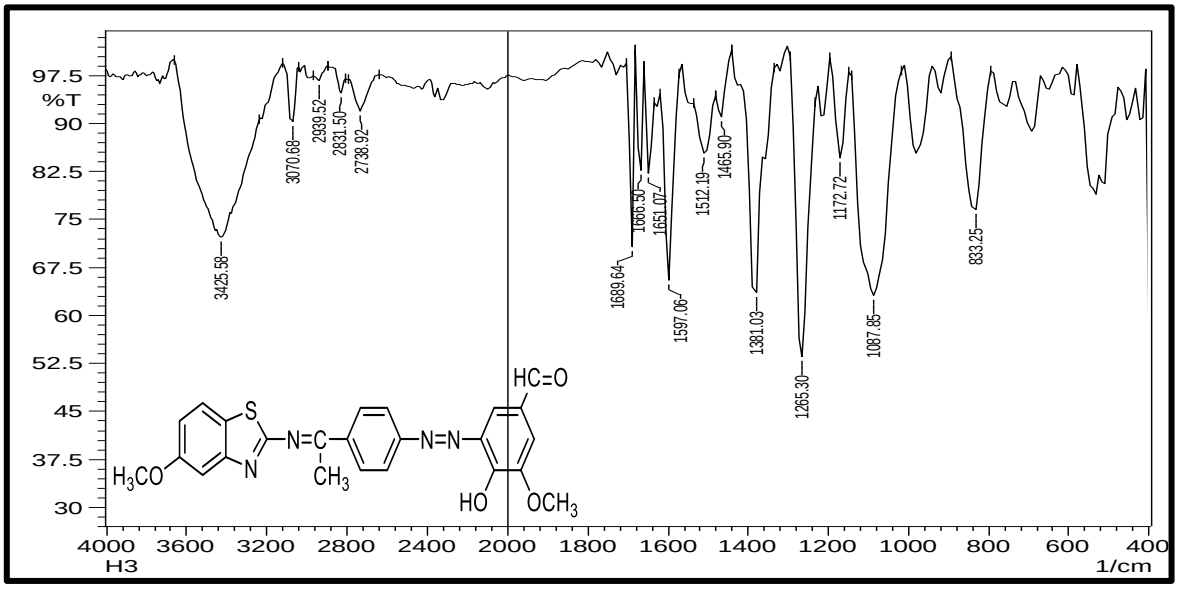


Scheme(3): Biological Activity Values of Compounds Prepared Against Four Types of Bacteria at 50 mg/mL

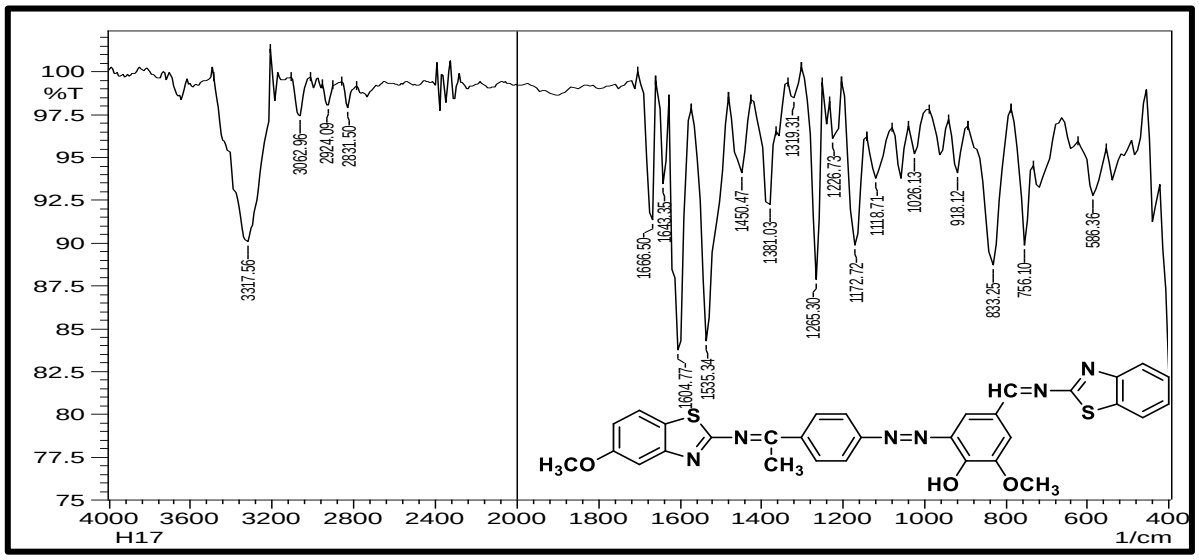


Figure(1): The FT-IR spectrum of compound [A]

ISSN: 0975-3583,0976-2833 VOL14, ISSUE 08, 2023

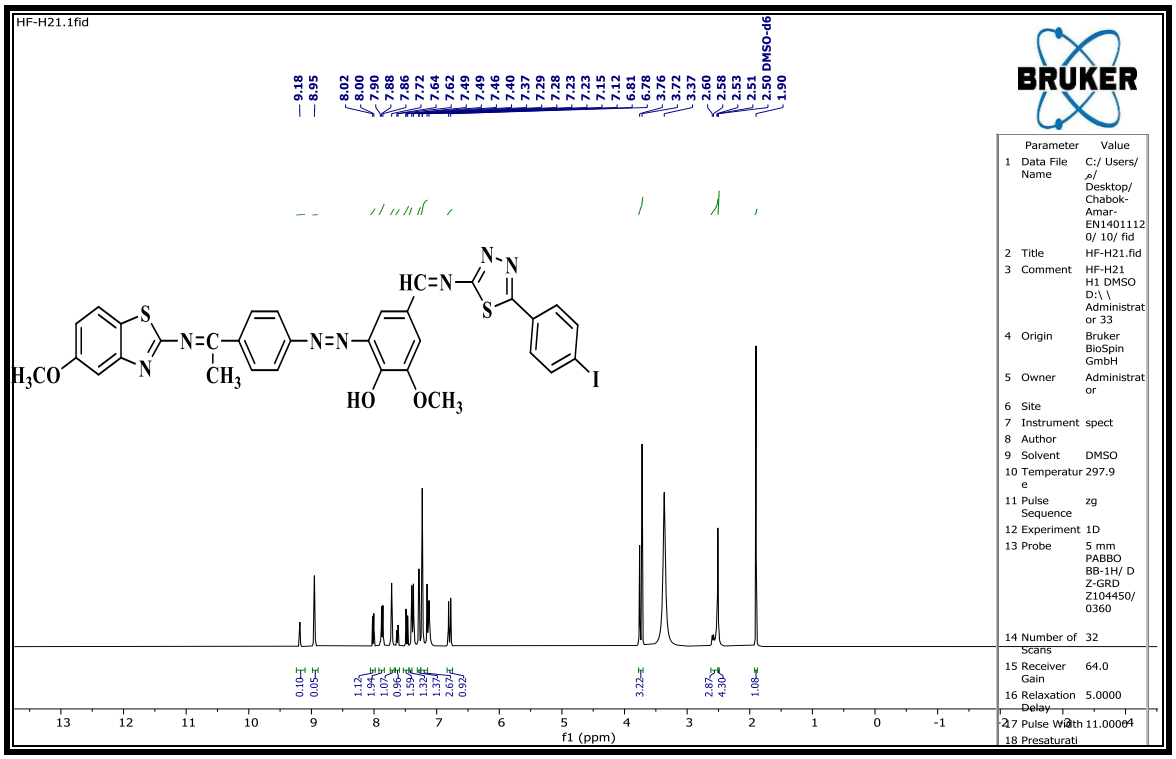


Figure(2): The FT-IR spectrum of compound [B]

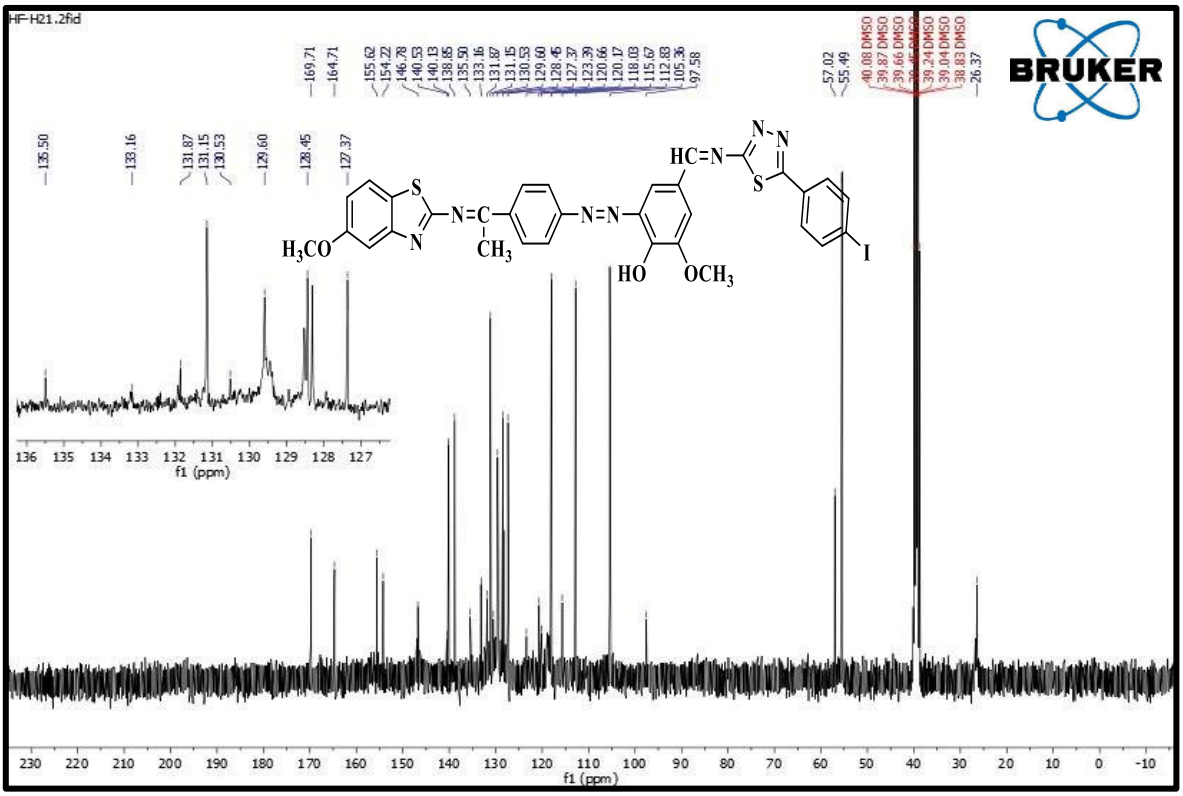


Figure(3): The FT-IR spectrum of compound [C]

ISSN: 0975-3583,0976-2833 VOL14, ISSUE 08, 2023



Figure(4): The ^{1}H-NMR spectrum of compound [C5]



Figure(5): The ^{13}C-NMR spectrum of compound [C5]

ISSN: 0975-3583,0976-2833 VOL14, ISSUE 08, 2023

Conclusion

In the present work, ten derivatives of azo compounds containing a 2-amino-6-methoxybenzothiazole ring were prepared, all compounds were characterization by spectral methods such as techniques FTIR, ^{1}H-NMR and ^{13}C-NMR, all compounds for biological activity, antibacterial activity was evaluated against four types of bacteria and the compounds showed moderate activity (*S. aureus and S. epidermidis*) and (*K. pneumonia and E. coli*) .

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ISSN: 0975-3583,0976-2833 VOL14, ISSUE 08, 2023

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