Article

Preparation, Characterization and Study of Color Stability with the Biological activity of Azo Dye Precursors and Schiff Bases Derived from Salicylaldehyde

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Abstract

In this study, derivatives of azo dyes (R1-R3) were prepared by preparing diazonium salt (prepared from the reaction of 4-aminoacetophenone or 3-aminopyridine with hydrochloric acid and sodium nitrite) and then coupling it with Salicylaldehyde dissolved in a basic solution, at a temperature of (0-5) ℃. Schiff base-azo derivatives (R4-R7) were synthesized by reacting equimolar amounts of the prepared azo dye compounds (R1, R2) with various substituted amines. Then, the accuracy of the structures of the prepared compounds were diagnosed and confirmed using physical and spectroscopic methods for the prepared compounds, such as infrared spectroscopy (FT-IR), proton and carbon nuclear magnetic resonance spectra (^{1}H,^{13}C-NMR). In addition to determining the melting points of the prepared compounds. The dyeing or color stability of azo dye derivatives (R1-R3) was studied in two steps. In the first step, the fibers are cleaned by removing impurities; dust stuck in them and shortening processes. Then, in the second step, the dyeing process is carried out by dissolving the dye. In the solvent 1.4-dioxane for dyeing pieces of cotton, wool, and wood. The biological activity of selected compounds was evaluated by studying their effects on the growth of two bacterial strains: ***Escherichia coli*** and ***Staphylococcus aureus***. ***Amoxicillin*** was used as a control antibiotic, and some of the prepared compounds exhibited significant inhibitory activity against the tested bacteria. The effect of some prepared compounds on the growth of one type of fungus, which is ***Candida albicans***, was studied. Antibiotics such as ***Fluconazole*** and ***Itraconazole*** were used as control samples, and some of the prepared compounds showed good inhibitory activity against the fungus used.

Keywords: Azo Dye, Schiff Base, Color Stability, Salicylaldehyde, Organic Synthesis, Bacterial Bioactivity, Fungal Biological.

1. Introduction

Azo dyes were discovered for the first time in 1888 AD by Peter [1]. They are organic substances that have the structure of the azo group [2]. Its colored compounds are absorbed in the visible and ultraviolet regions [3]. These compounds are used in Coloring other materials by bonding with them, giving them stable colors that are not affected by washing, acids, bases, or oxygen [4]. There are two types of dyes: natural dyes and artificial dyes [5]. The natural ones are extracted from animals and plants, and the artificial ones are prepared using aniline, which is considered the primary material for their preparation [6]. Azo dyes are linked to aromatic or aliphatic groups, as aromatic azo dyes have gained widespread due to their high stability [7]. The

reason for this stability is due to the type of groups attached to the azo group and the number of those groups, as the number of successive double bonds increases with the double bond of the azo group [8]. Azo leads to improved stability due to the occurrence of the resonance phenomenon [9]. Azo dyes have been used in purification processes, as well as withdrawing heavy metal ions from polluted water by adsorption method by forming complexes with activated charcoal [10]. Azo dyes have also been used as indicators in dyeing processes [11]. They are used as reagents to measure optical intensity, as well as in the processes of dyeing cotton, silk, wood, and wool. An example of this is the dye (β-Naphthol orange) used to dye wool and cotton [12]. Diazonium salts are the primary compound for preparing azo dyes, as diazonium salts can combine with many compounds that have high electronic density because diazonium salts have electrophilic properties [13]. Diazonium salts can be obtained through an azo reaction, which is done by treating Nitrite ions with the primary aromatic amine in an acidic environment at (0-5) °C [14]. Azo dyes are formed by a conjugation reaction, characterized by its simplicity and speed [15]. Conjugation occurs between two compounds, one of which is an aromatic amine or phenol, and the other is a diazonium salt [16]. They are those compounds that contain the azomethine group and are often distinguished by their yellow color [17]. They were given this name after the chemist Hugo Schiff [18]. They are imines in which the nitrogen atom is linked to an aryl or alkyl but not to a hydrogen atom so the association of the organic part with the nitrogen atom has a role in increasing the stability of the resulting imine and preserving it from decomposition or polymerization [19, 20]. Schiff bases are a class of compounds that have been studied because of their effectiveness, physical properties [21], and many applications in several fields, the most important of which are industrial fields, as they were used as corrosion inhibitors and catalysts and in the preparation of polymers, as they were used in polymers to increase the electrical conductivity property and they are used as complexes Schiff bases as oxidizing agents [22]. Many Schiff base complexes show distinct catalytic activities in industrial applications such as oxidation and redox processes [23], as they can serve as effective photoactive materials and optical materials. Schiff bases are also used in the manufacture of dyes [24]. It was also proven that some Schiff bases have medicinal properties such as heart tonics and diuretics and Schiff bases showed clear effectiveness against leukemia and brain cancer [25].

2. Experimental:

2.1. Material and Devices used: All chemicals used in this work were purchased from Fluka, Aldrich, and BDH and used without further purification.The melting points were measured using Electrothermal Melting Apparatus 9300. Shimadzu FT-IR 8400S spectrophotometer with a scale of (400-4000) $cm^{-1}$by KBr disc.^{1}H-NMR and ^{13}C-NMR spectra on Bruker instruments running at 400 MH_Z. Thin Layer Chromatography (TLC) was performed using silica gel plates with 0.2 mm thickness, activated with fluorescent silica gel, and visualization was achieved using UV light.

2.2. Preparation of azo dye derivatives (R1-R3) [26, 27]

The first step: Preparation of diazonium salt: Dissolve (0.01 mol) of each of the primary amine substitutes (3-aminopyridine, 4-aminoacetophenone, 4-(phenyldiazine) aniline) in (30 ml) an acidic solution of 37% hydrochloric acid. (20 ml distilled water + 10 ml hydrochloric acid) at a temperature of (0-5) ℃ and place the mixture in a conical flask with continuous stirring. Then, in a second conical flask, dissolved (0.01 mol, 0.68 grams) of sodium nitrite ($NaNO_2$) in the smallest possible amount of iced distilled water (2 ml) and added it to the solution of the first flask, with continuous stirring, and the addition was gradual and continuous. It forms droplets, as a color

change was observed, and this is evidence of the formation of diazonium salt, which was stored at a temperature of (0-5) °C.

The second step: Preparing the coupling solution: dissolved (0.01 mol, 1 ml) of salicylaldehyde in (30 ml) of a basic solution at a temperature of (0-5) °C, and after completing the dissolution by continuous stirring, the diazonium salt prepared in the first step was added to the coupling solution at (0-5) °C with continuous stirring. After that, the product was poured onto ice grits, then filtered, dried at a temperature of (45 °C), and recrystallized with absolute ethanol. As Table (1) shows some of the physical properties of azo dyes, and as in scheme (1).

2.3. Preparation of derivatives of Schiff-azo bases (R4-R7) [28, 29]

(0.005 mol) of the prepared azo dye compound (R1) was dissolved in (30 ml) of absolute ethanol, and after complete dissolution, four drops of glacial acetic acid were added, then (0.005 mol) of various amine substitutes were added (phenylhydrazine, sulfanilamide, Amperon, sulfamethaxazole), and the mixture was stirred in reverse for (4-5) hours, and the completion of the reaction was confirmed using the thin layer chromatography technique. After the end of the reaction, the resulting mixture was slowly cooled, filtered, the precipitate was collected, dried until the weight was constant, and it was recrystallized with benzene and the characteristics the physics of compounds (R4-R7) is shown in Table (1).

2.4. Study of the color stability of azo dye derivatives (R1-R3)

The method of dyeing or studying the color stability of azo dye is usually carried out either on a continuous basis or in batches. The fibers are dyed either in the form of raw material or in the form of yarn or textile threads. The dyeing process requires the transfer of the dye from a solution Dyeing into the fabric [30], and the fundamental processes of dyeing include the following steps:

1. Preparing the fibers: The cleaning process includes the removal of impurities, fatty materials, and dust from natural fibers. As for manufactured fibers such as polyamides (nylon), they are cleaned by spinning and finishing processes [31].

2. Dyeing: Derivatives of azo dyes (R1-R3) were used in dyeing processes. (0.1 g) of the azo dye was dissolved in (25 ml) of the solvent 1,4-Dioxane, and the solutions were used. The result of dyeing pieces of equal weight (200 mg) of cotton, wool, and wood. Each type of these materials was placed in the beaker containing the dye (total immersion) with stirring for (30 minutes), and then the beaker was left in the oven, at a temperature of (100 °C) and to the point of dryness [32].

2.5. Evaluation of bacterial and fungal bioactivity [33, 34]

Two types of bacterial isolates were tested, one was gram-negative [Gr-ve], which is ***Escherichia coli***, and one was gram-positive [Gr+ve], which is ***Staphylococcus aureus***. One type of fungal isolate was tested, which is ***Candida albicans***. The Agar-well diffusion method was followed to test the effectiveness of the prepared compounds (R1, R3, R5, R6, R7) on bacteria. Inoculating the culture medium with bacterial isolates, holes were made in the dishes using the Cylinder metric method using a cork drill with a diameter of (6 mm), and (20-100) microliters of the compounds prepared for the three concentrations were placed in each hole from the pits, then the dishes were incubated in the incubator at a temperature of (37 °C) [35]. The results were read after (24-48) hours for bacterial and (7-14) days for fungal to show the sensitivity of the derivatives used, which depends on the diameter of inhibition evident in the dishes around the holes used, as the increase in the diameter of inhibition means an increase in the biological effectiveness of the prepared compounds and comparing that with the diameter of inhibition of standard antibiotics.

Standard antibiotics were used in solutions such as ***Amoxicillin***, ***Ampicillin***, ***Ciprofloxacin,*** ***Fluconazole*** and ***Itraconazole*** as control samples [36].

3. Results and discussion

3.1. Characterization of Azo dye derivatives (R1-R3)

3.1.1. Mechanism of preparing Azo dye derivatives (R1-R3)

Azo dye derivatives (R3-R1) were prepared by reacting equal moles of primary amine substitutes (3-aminopyridine, 4-aminoacetophenone, 4-(phenyldiazine) aniline) dissolved in a nitrous acid solution (sodium nitrite and hydrochloric acid). Diluted 37%), with series aldehyde dissolved in a basic solution (ethanol and sodium hydroxide). The mechanism of preparing azo dyes is summarized in many steps: In the first step, nitrous acid is prepared through the reaction of sodium nitrite and dilute hydrochloric acid (37%), and in the second step, the electron pair in the amine group is attacked on the group (N=O) in nitrous acid, and a bond occurs between the two groups. Then, in the third step, the two protons attached to the amine group are released, followed by the addition of a proton and chlorine from hydrochloric acid. In this step, the oxonium ion is formed. In the fourth step, a water molecule is released to form the diazonium salt, which is unstable at average temperatures, so it is kept at a temperature of (0-5) degrees Celsius. In the fifth step, the coupling solution consisting of salicylaldehyde dissolved in a basic solution is added, where it is formed in the negative carbonation ion. In the sixth and final step, the connection between the diazonium salt and the negative carbonation ion to form the azo dye, as in the scheme (2), which shows the known mechanism [37] for preparing azo dyes (R1-R3).

3.1.2. Characterization of Azo dye derivatives (R1-R3) by FT-IR

When studying the infrared spectrum of the azo dye derivative (R1), it was observed that the (NH_2) band of the amine derivatives disappeared, and the appearance of a new absorption band at a frequency (3417) cm^{-1} that belongs to the stretching of the hydroxyl bond (OH). The appearance of an absorption band at the frequency (3055) cm^{-1} is due to the stretching of the aromatic (CH) chain. The appearance of an absorption band at the frequency (1720) cm^{-1} due to the stretching of the carbonyl aldehyde (C=O). It was also observed that two absorption bands appeared at frequencies (1597) cm^{-1} and (1485) cm^{-1} due to the stretching of the aromatic (C=C) bond [38, 39], with the appearance of an absorption band at frequency (1438) cm^{-1} due to stretching (N=N), as shown in Figure (1).

When studying the infrared spectrum of the azo dye derivative (R2), it was observed that the (NH_2) band of the amine derivative disappeared, and the appearance of a new absorption band at the frequency (3363) cm^{-1} that belongs to the stretching of the hydroxyl bond (OH). The appearance of an absorption band at frequency (3051) cm^{-1}, which is due to the stretching of the aromatic (CH) sphincter, and the appearance of two absorption bands at frequencies (2924) cm^{-1} and (2866) cm^{-1}, which is due to the stretching of the (CH_3) sphincter.), and the appearance of an absorption band at a frequency (1735) cm^{-1} due to the stretching of the aldehyde carbonyl (C=O), and the appearance of an absorption band at frequency (1724) cm^{-1} due to the stretching of the ketone carbonyl (C=O), it was also observed that two absorption bands appeared at frequencies (1593) cm^{-1} and (1485) cm^{-1} due to the stretching of the aromatic bond (C=C), with an absorption band appearing at frequency (1435) cm^{-1}due to the stretching of the sphincter (N=N) [40], as shown in Figure (2).

When studying the infrared spectrum of the azo dye derivative (R3), it was observed that the (NH_2) band of the amine derivatives disappeared, and the appearance of a new absorption band at frequency (3309) cm^{-1} which belongs to the stretching of the hydroxyl bond (OH). The appearance

of an absorption band at the frequency (3063) cm^{-1} which is due to the stretching of the aromatic (CH) bond, and the appearance of an absorption band at the frequency (1724) cm^{-1} which is due to the stretching of the carbonyl aldehyde bond (C=O), It was also observed that two absorption bands appeared at frequencies (1597) cm^{-1} and (1489) cm^{-1} due to the stretching of the aromatic (C=C) conjugate, with the appearance of an absorption band at frequency (1435) cm^{-1} due to the stretching of the (N=N) group [41], as shown in Figure (3).

3.1.3. Characterization of Azo dye derivatives (R1-R3) by NMR

When studying the ^{1}H-NMR spectrum of the proton of the compound (R2) using a solvent (DMSO-d^6), it was observed that a single signal appeared at δ (10.77) ppm, attributed to a proton. The hydroxyl group (OH) (b), and the appearance of a single signal at δ (10.26) ppm due to the protonation of the aldehyde group (HC=O) (a), and the appearance of a multiple signal in The range is (6.57-8.23) ppm attributed to the protons of the aromatic rings (c, d, e, f, g), and the appearance of a signal at δ (3.45 and 3.47) ppm attributed to (HDO), and the appearance of a signal at δ (2.50-2.52) ppm attributed to the protons of the solvent ((DMSO-d^6), and the appearance of a single signal at δ (2.38) ppm attributed to the proton of the group (CH_3) (h) [42], and as in Figure (4).

When studying the 13C-NMR spectrum of carbon of the compound (R2) using a solvent (DMSO-d^6), it was observed that a single signal appeared at δ (195.48) ppm, attributed to carbon. The carbonyl group (C=O) (i), and the appearance of a single signal at δ (192.12) ppm attributed to the carbon of the carbonyl group (C=O) (a), and the appearance of a single signal at δ (145.29) ppm is attributed to the carbon of the (C-OH) group (b). The appearance of a multiple signal at δ (113.08-138.45) ppm is due to the carbons of the aromatic ring (c, d, e, f, g, h), and the appearance of a signal at δ (39.30-40.55) ppm attributed to the carbon of the solvent (DMSO-d^6), and the appearance of a single signal at δ (27.21) ppm are attributed to the carbon of the two groups (CH_3) (j) [43], and the spectrum is shown in Figures (5).

3.2. Characterization of Schiff-Azo base derivatives (R4-R7)

3.2.1. Mechanism of Schiff-Azo base derivatives (R4-R7)

The mechanism of preparing derivatives of Azo -Schiff bases (R4-R7) is summarized in two steps: addition and deletion: in the first step, the unshared electron pair of the nitrogen atom in the amine group is attacked by the carbon atom in the carbonyl group in the aldehyde, while in the second step, deletion of the water molecule occurs. To form the azomethine bond (C=N), drops of glacial acetic acid or hydrochloric acid are added for the purpose of protonating the carbonyl oxygen and making the carbon atom more electrophilic [44].

3.1.2. Characterization of Schiff-Azo base derivatives (R4-R7) by FT-IR

When studying the infrared spectrum of the Azo -Schiff bases (derivatives (R4-R7), it was observed that the aldehyde carbonyl band of the prepared azo dye derivatives disappeared, as well as the disappearance of the amine group (NH_2) of the amines used in the preparation of the Azo -Schiff bases derivatives (R4-R7), and the appearance of an absorption band. New in the range (1627-1639) cm^{-1} due to the stretching of the azomethine bond (HC=N), and the appearance of an absorption band in the range (3360-3433) cm^{-1} due to the stretching of the hydroxyl (OH) bond, and the appearance of an absorption band in the range (3051- 3089) cm^{-1} is due to the stretching of the aromatic (CH) bond. It was also observed that two absorption bands appeared in the range (1589-1598) cm^{-1} and (1482-1497) cm^{-1} due to the stretching of the aromatic (C=C) bond, and the

appearance of an absorption band. In the range (1425-1438) cm^{-1}, it returns to the elastic stretch (N=N) [45], as shown in Table (2) and Figures (6, 7).

3.1.3. Characterization of Azo -Schiff bases **derivatives (R4-R7) by NMR**

When studying the ^{1}H-NMR spectrum of the compound (R4) using solvent (DMSO-d^6), it was observed that a single signal appeared at δ (10.57) ppm attributed to the proton of the hydroxyl group (OH) (h), and the appearance of a signal Single at δ (10.49) ppm attributed to the proton of the (NH) group (j), and the appearance of a single signal at δ (8.17) ppm attributed to the proton of the azomethine group (HC=N) (i), and the appearance of a multiple signal In the range (7.26-7.65) ppm is attributed to the protons of the aromatic rings (a, b, c, d, e, f, g, k, l, m), and a signal appears at δ (3.54 and 3.60) part of ppm is attributed to (HDO), and the appearance of a signal at δ (2.51) ppm is attributed to the solvent (DMSO-d^6) [46], as in Figure (8).

When studying the ^{1}H-NMR spectrum of compound (R5) using solvent (DMSO-d^6), it was observed that a single signal appeared at δ (10.77) ppm attributed to the protonation of the hydroxyl group (OH) (h), and the appearance of a signal Single at δ (7.93) ppm attributed to the proton of the azomethine group (HC=N) (i), and the appearance of a multiple signal in the range (7.50-7.71) ppm attributed to the protons of the aromatic rings (a, b, c, d). ,e,f,g,j,k), and the appearance of a single signal at δ (7.45) ppm attributed to the proton of the (NH_2) group (l), and the appearance of a signal at δ (3.49) ppm attributed to (HDO), and the appearance of a signal at δ (2.50) ppm attributed to the solvent (DMSO-d^6) [47], as in Figure (9).

When studying the ^{1}H-NMR spectrum of compound (R6) using solvent (DMSO-d^6), it was observed that a single signal appeared at δ (12.49) ppm attributed to the proton of the (NH) group (l). At δ (10.76) ppm is attributed to the proton of the hydroxyl group (OH) (h), and a single signal appears. At δ (8.96) ppm is attributed to the proton of the azomethine group (HC=N) (i), and a multiple signal appears In the range (7.47-7.70) ppm is attributed to the protons of the aromatic rings (a, b, c, d, e, f, g, j, k), and a single signal appears at δ (6.11) ppm is attributed to a proton. (HC=C) (m) group in the oxazole ring, and a signal appears at δ (3.46) ppm due to (HDO), and a signal appears at δ (2.50) ppm due to the protons of the solvent (DMSO-d^6), and the appearance of a single signal at δ (2.30) ppm, attributed to the proton of the methyl group (CH_3) (n) [48], as shown in Figure (10).

When studying the ^{1}H-NMR spectrum of compound (R7) using solvent (DMSO-d^6), it was observed that a single signal appeared at δ (10.50) ppm attributed to the protonation of the hydroxyl group (OH) (h), and the appearance of a signal Single at δ (8.69 ppm) attributed to the proton of the azomethine group (HC=N) (i), and the appearance of a multiple signal in the range (7.24-7.55) ppm attributed to the protons of the aromatic rings (a, b,c,d,,e,f,g,l,m,n), and the appearance of a signal at δ (3.41) ppm is attributed to (HDO), and the appearance of a single signal at δ (3.21) ppm is attributed to the proton of the methyl group (CH_3) (k), a signal appears at δ (2.50) ppm due to the protons of the solvent (DMSO-d^6), and a single signal appears at δ (2.45) ppm due to the proton of the methyl group (CH_3) (j), and as in Figure (11).

When studying the ^{13}C-NMR spectrum of carbon (R7), it was observed that a signal appeared at δ (159.88) ppm attributed to the carbon of the azomethine group (HC=N) (k), and a signal appeared at δ (157.92) ppm attributed to the carbon of the carbonyl group (C=O) (n), a signal at δ (150.66) ppm attributed to the carbon of the group (C-OH) (h), and a multiple signal at δ (119.68-134.64) ppm It belongs to the carbons of the aromatic rings (a,b,c,d,e,f,g,i,j,q,r,s,t), and the signal at δ (116.87) ppm is attributed to the carbon of the group (C= C) (m), a signal at δ (114.46) ppm is attributed to the carbon of the group (C=C) (l), and a signal at δ (39.30-40.55) ppm is attributed to the carbon of the solvent (DMSO-d^6) A single signal at δ (35.60) ppm is attributed to

the carbon of the two groups (CH_3) (p), and a single signal at δ (10.32) ppm is attributed to the carbon of the two groups (CH_3) (o) [49], and as in Figures (12).

3.3. Study of the color stability of azo dye derivatives (R1-R3)

The prepared azo dye compounds (R1-R3) were tested in the dyeing process using three materials: cotton, wool, and wood, according to the dyeing process in the practical part, the results showed solid and transparent stability after washing with water, as well as strong stability after washing with liquid soap. The most stable compound is the compound (R3). This is due to the high polarity of the compound, as well as the higher ability of wood to form hydrogen bonds with the dyes used in the study [50], as shown in Figures (13-15).

3.4. Evaluation of the bacterial and fungus biological activity

The biological effectiveness of the prepared compounds (R1, R3, R5, R6, R7) was evaluated in this study on two types of bacteria, namely ***Escherichia coli,Staphylococcus aureus*** and fungus ***Candida albicans***. These bacteria and fungus were chosen due to their medical importance, as they cause many diseases [51]. In addition, they differ in their resistance to antibiotics. The biological effectiveness of the prepared compounds was evaluated using the etching method and measuring the level of inhibition (Inhibition zone) [52]. The results indicate that the compounds The preparation could inhibit the growth of bacteria used in both types, positive and negative for the gram stain, in varying proportions [53]. The antibiotics ***Amoxicillin***, ***Ampicillin***, ***Ciprofloxacin***and ***Fluconazole*** were used as control samples, based on what is used in the laboratories of the Ministry of Health and based on World Health Organization tests, as the compounds used in the study of biological effectiveness against ***E. coli*** bacteria showed no inhibition at the concentration (25 mg/ml) and the concentration (50 mg/ml), and showed good inhibitory activity at the concentration (100 mg/ml). Likewise, the compounds used in the study of biological effectiveness against ***S. aureus*** bacteria did not show any inhibition. At the concentration (25 mg/ml) and the concentration (50 mg/ml), it showed good inhibitory effectiveness at the concentration (100 mg/ml) [54], as in Table (3) and Figure (16).

Table (1): Some physical properties of prepared azo dye derivatives (R3-R1).

Comp. No.	Molecular Formula/ M.Wt g/mol	Color	Time (h)	M.P (^{0}C)	Yield (%)
R1	$C_{12}H_9N_3O_2$ / 227.22	Brown	-----	150-152	75
R2	$C_{15}H_{12}N_2O_3$ / 268.27	Orange	-----	127-129	80
R3	$C_{19}H_{14}N_4O_2$ / 330.35	Dark red	-----	200-201	78
R4	$C_{18}H_{15}N_5O$ / 317.35	Dark red	5	115-116	69
R5	$C_{18}H_{15}N_5O_3S$ / 381.41	Yellow	5	220-222	65
R6	$C_{22}H_{18}N_6O_4S$ / 462.48	Orange	4	204-205	71
R7	$C_{23}H_{20}N_6O_2$ / 412.45	Dark yellow	5	187-189	76

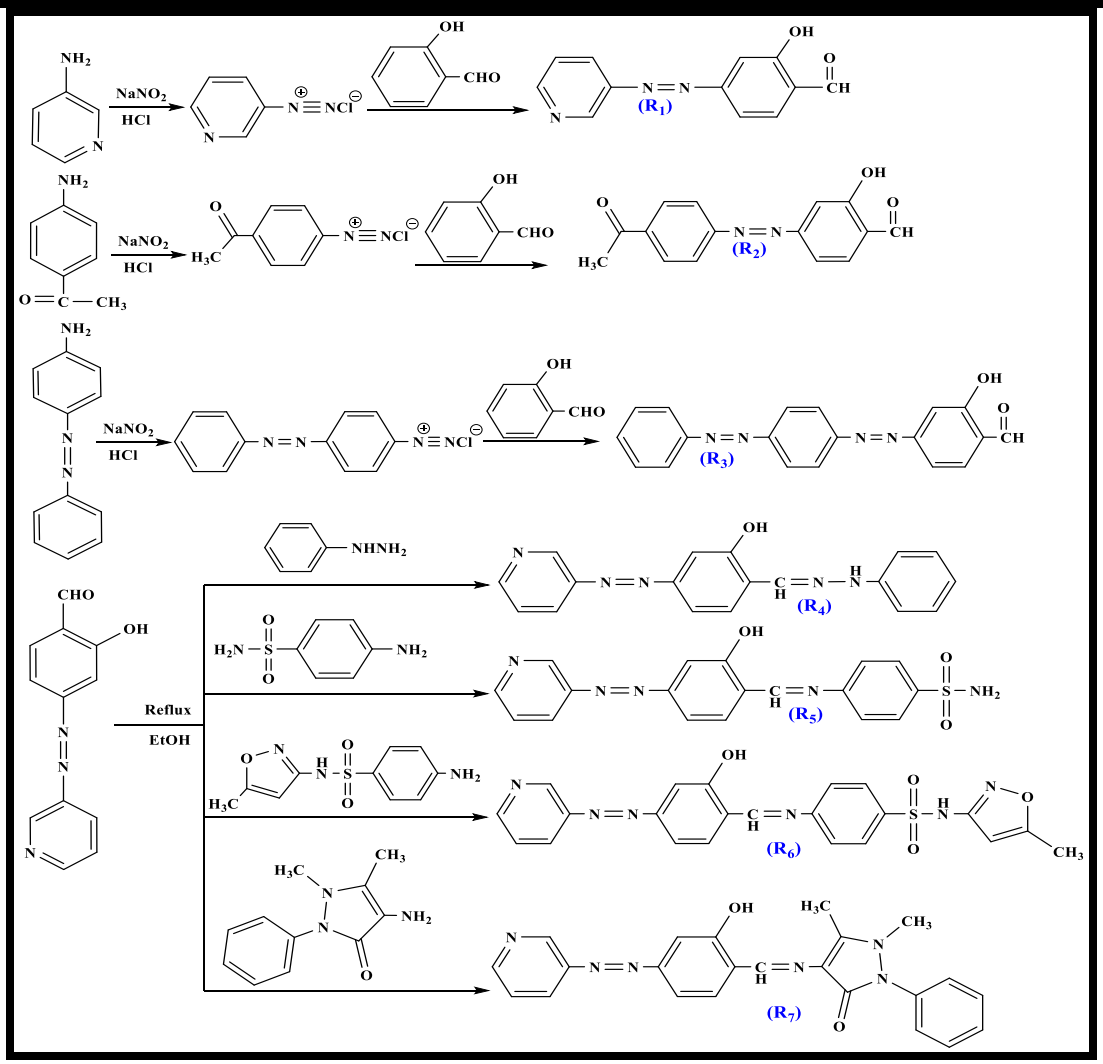
Table (2): Infrared absorption results (cm^{-1}) for of Schiff-Azo base derivatives (R4-R7).

Comp. No.	νO-H	νC-H Arom.	ν HC=N	νC=C arom.	νN=N	Others
R4	3421	3051	1639	1597, 1485	1427	ν (NH) 3182
R5	3390	3075	1638	1598, 1482	1430	ν (NH_2) 3302, 3210
R6	3412	3089	1636	1596, 1497	1425	ν (NH) 3191, and ν (CH_3) 2922, 2845

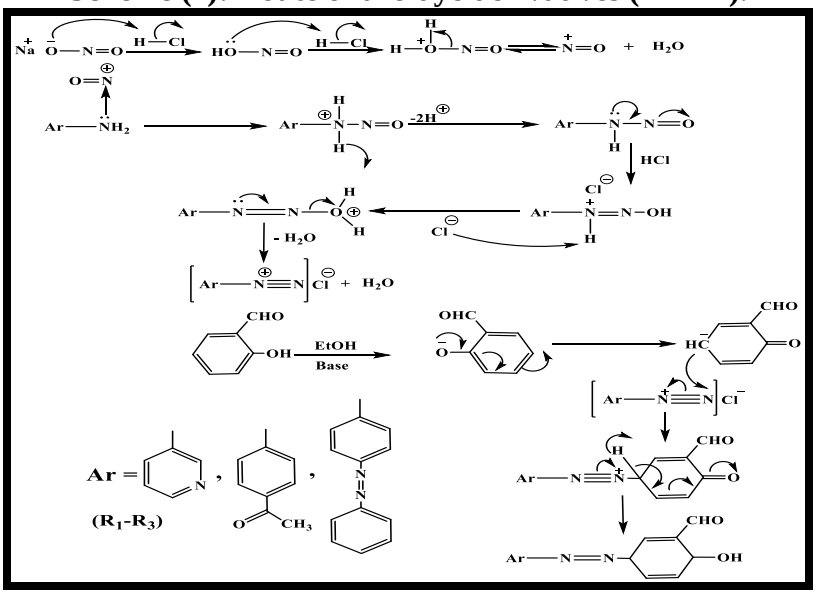
R7	3360	3066	1635	1589, 1489	1431	ν (CH_3) 2993, 2889, and ν (C=O) 1705

Table (3): Biological effectiveness of the prepared compounds and antibiotics (mm).

Comp. No.	***Escherichia coil***			***Staphylococcus aureus***			***Candida albicans***		
Conc. mg/ml	**25**	**50**	**100**	**25**	**50**	**100**	**25**	**50**	**100**
R1	0	0	23	0	0	21	0	0	35
R3	0	0	25	0	0	26	0	0	10
R5	0	0	20	0	0	0	0	0	40
R6	0	0	10	0	0	22	0	0	5
R7	0	0	0	0	0	0	0	0	0
Amoxicillin	14	17	24	10	14	25	---	---	---
Ampicillin	10	16	22	14	20	24	---	---	---
Ciprofloxacin	12	17	21	15	18	21	---	---	---
Fluconazole	---	---	---	---	---	---	17	28	39
Itraconazole	---	---	---	---	---	---	22	30	40
Blank disk	0	0	0	0	0	0	0	0	0



Scheme (1): Route of azo dye derivatives (R1-R7).



Scheme (2): Mechanism of preparing azo dye derivatives (R1-R3).

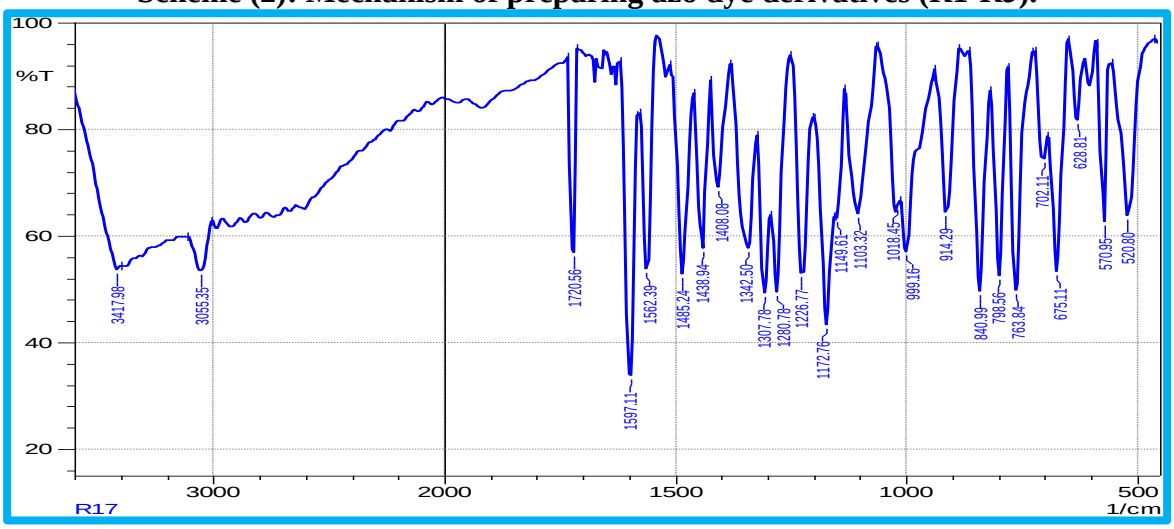


Figure (1): FT-IR spectrum of the compound (R1).

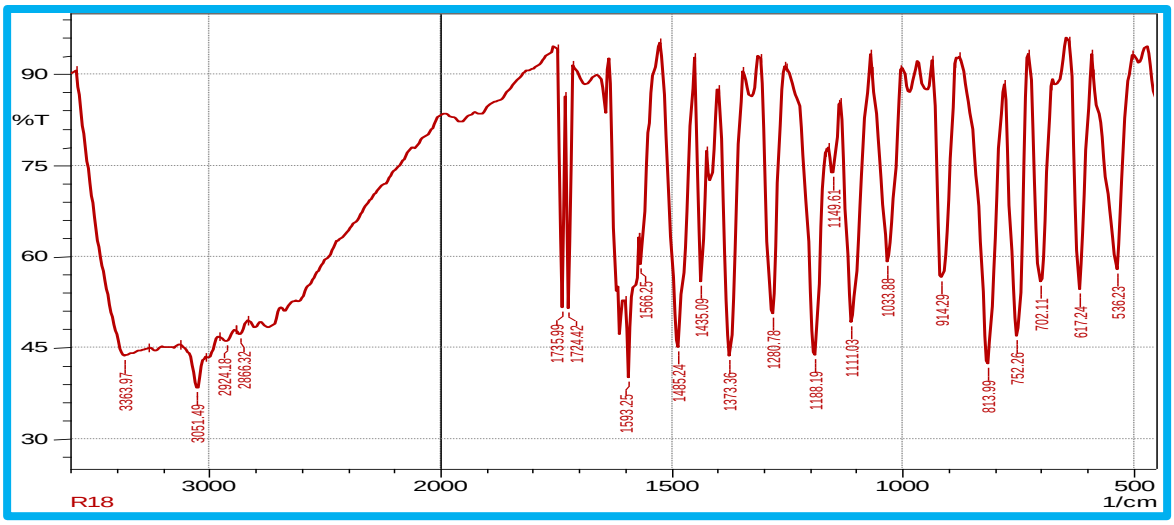


Figure (2): FT-IR spectrum of the compound (R2).

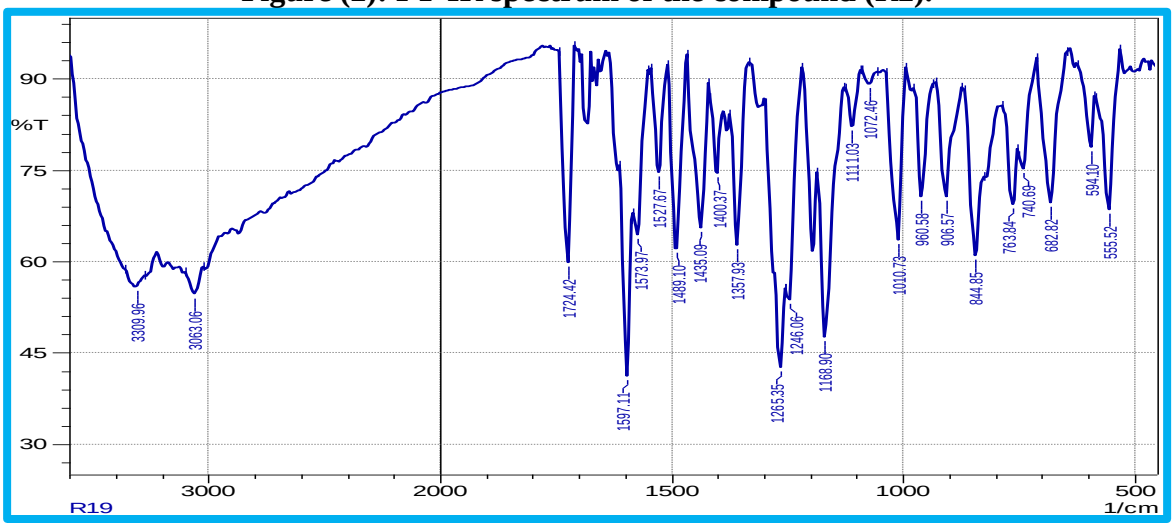


Figure (3): FT-IR spectrum of the compound (R3).

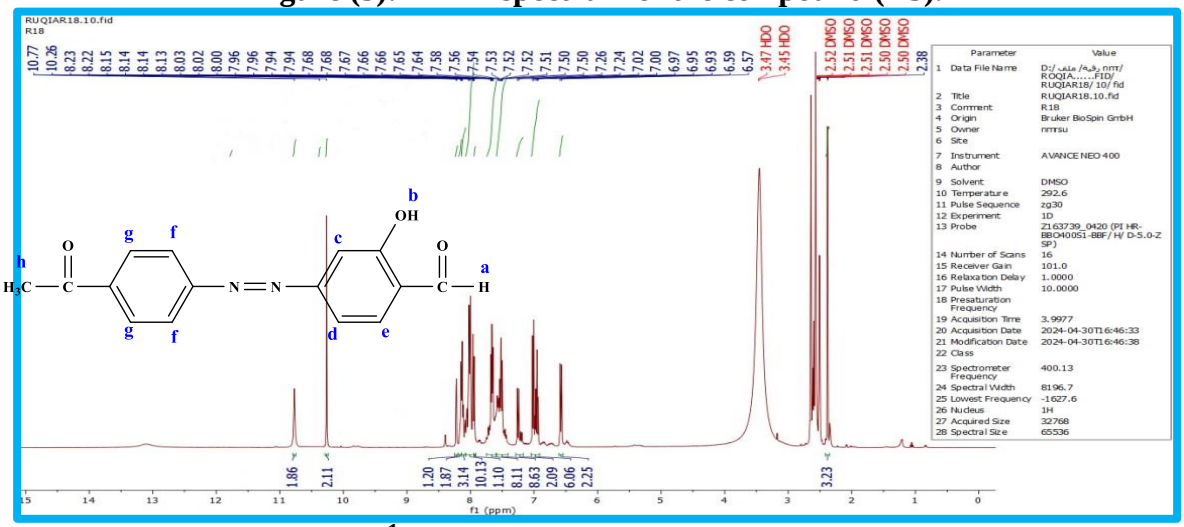


Figure (4): ^{1}H-NMR spectrum of the compound (R2).

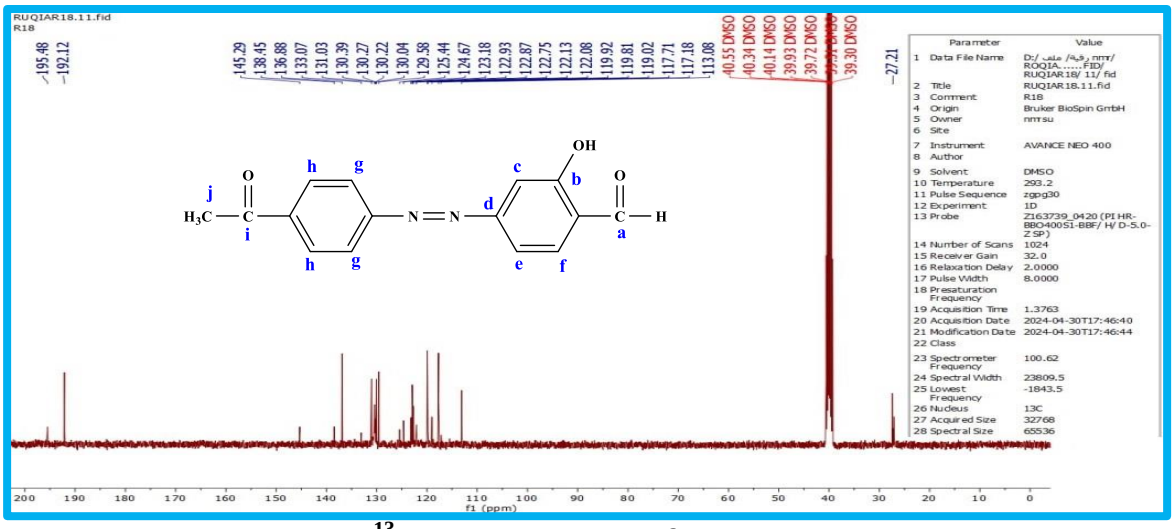


Figure (5): ^{13}C-NMR spectrum of the compound (R2).

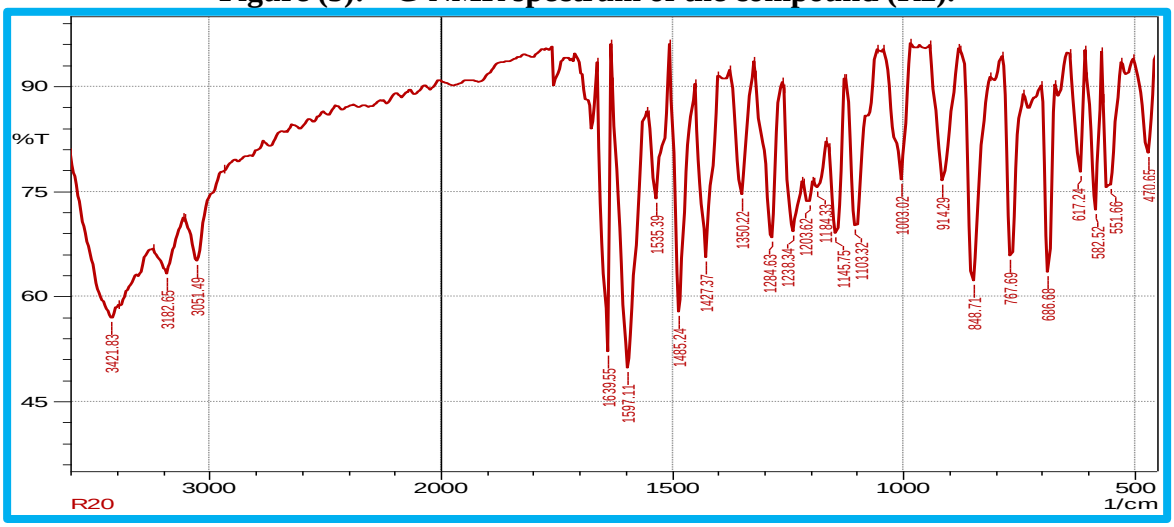


Figure (6): FT-IR spectrum of the compound (R4).

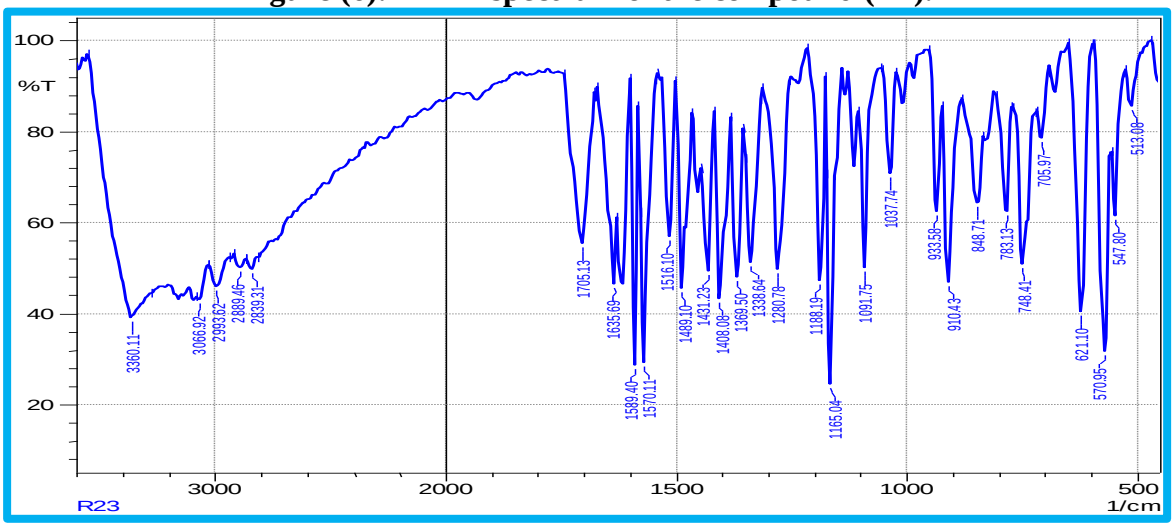


Figure (7): FT-IR spectrum of the compound (R7).

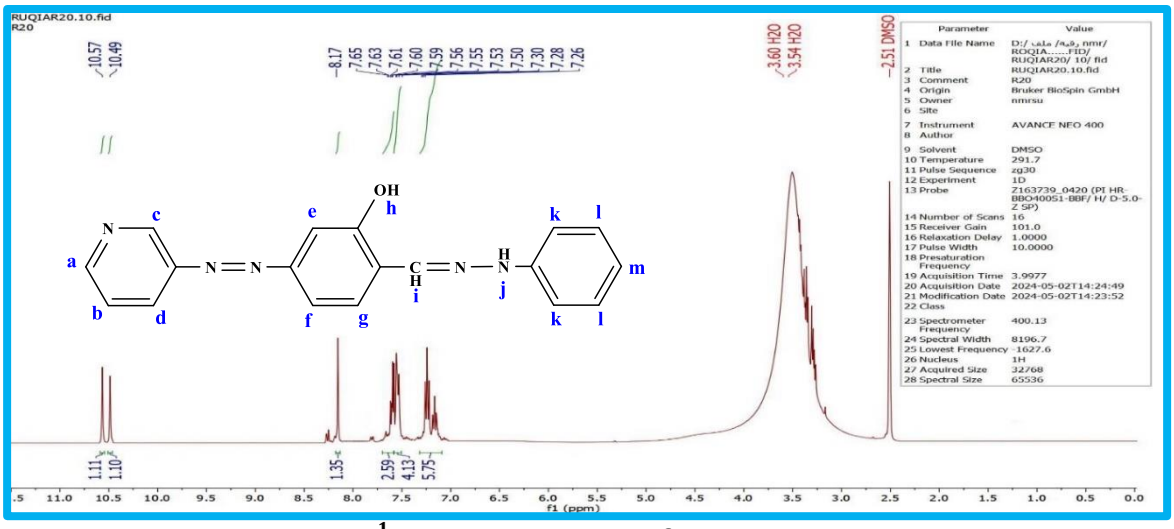


Figure (8): ^{1}H-NMR spectrum of the compound (R4).

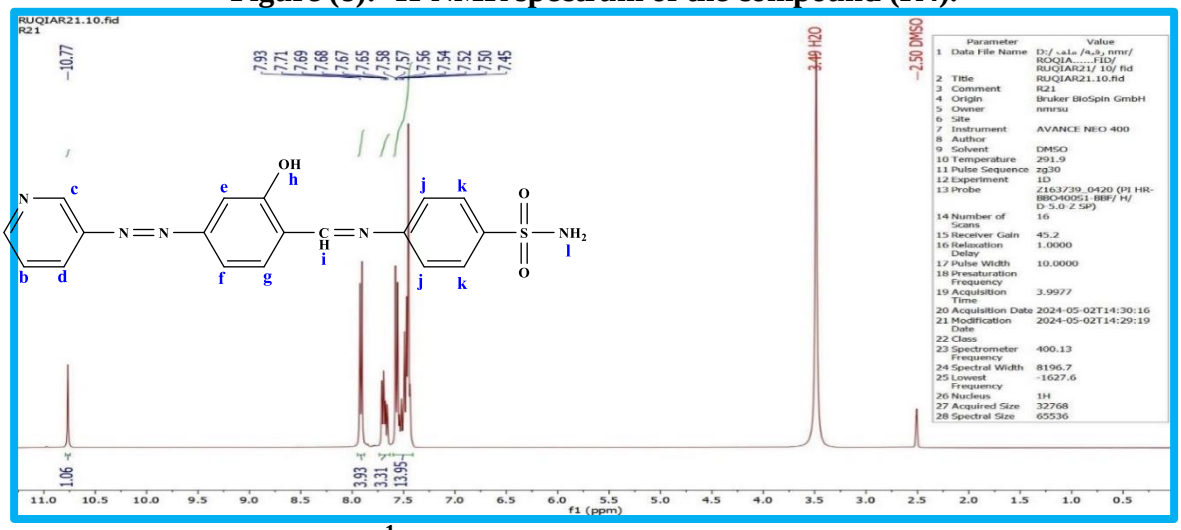


Figure (9): ^{1}H-NMR spectrum of the compound (R5).

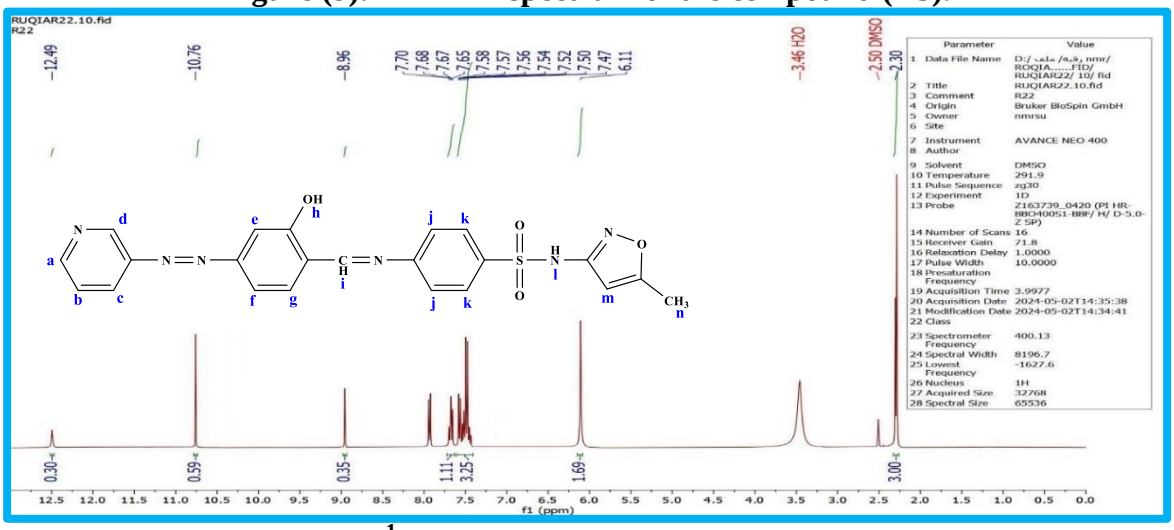


Figure (10): ^{1}H-NMR spectrum of the compound (R6).

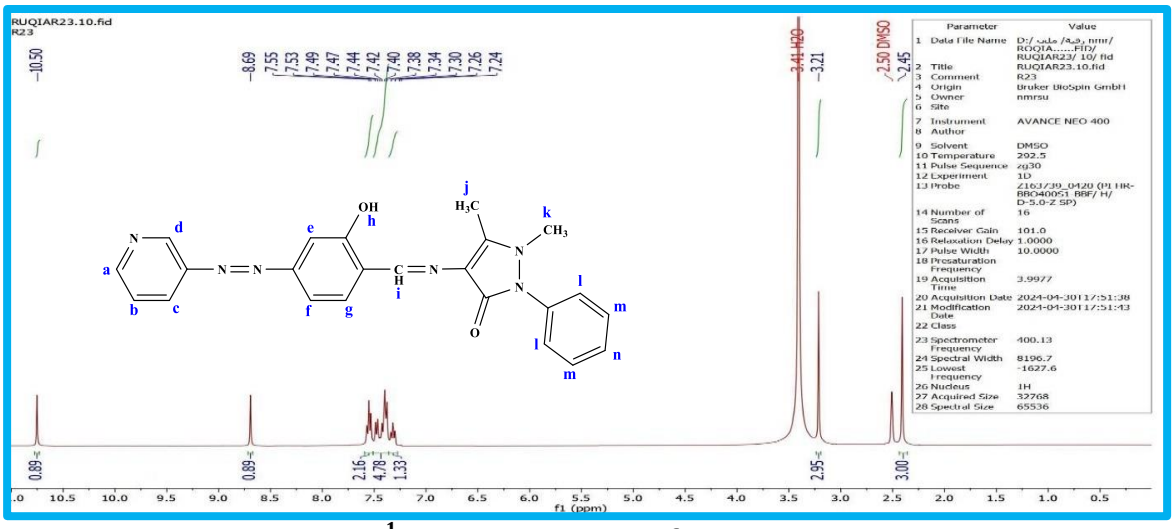


Figure (11): ^{1}H-NMR spectrum of the compound (R7).

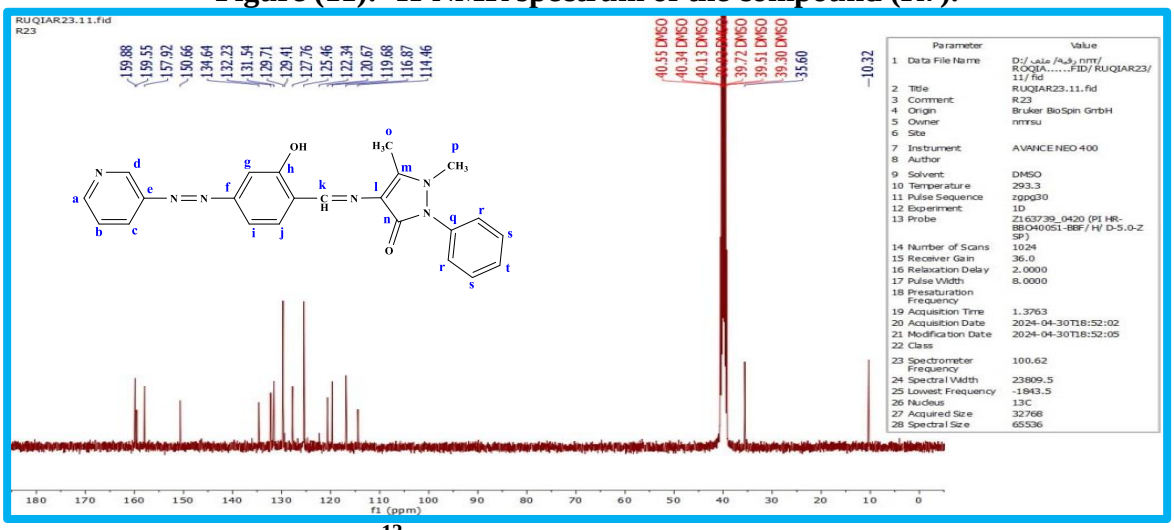


Figure (12): ^{13}C-NMR spectrum of the compound (R7).

Figure (13): Fabrics dyed with compound (R1).

Figure (14): Fabrics dyed with compound (R2).

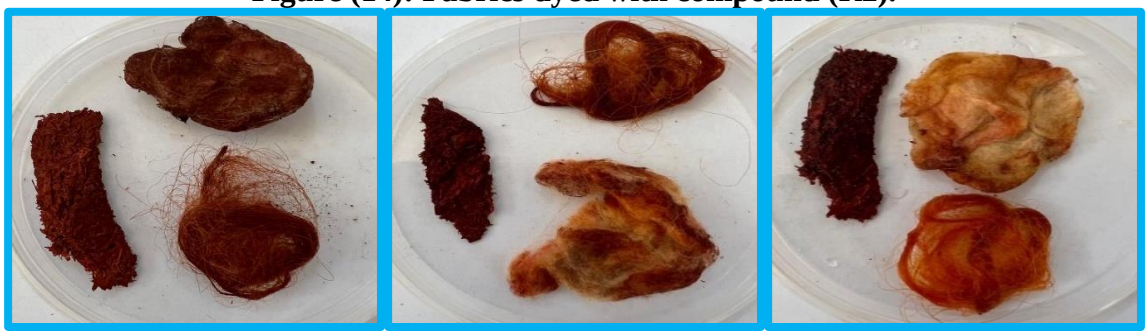


Figure (15): Fabrics dyed with compound (R3).

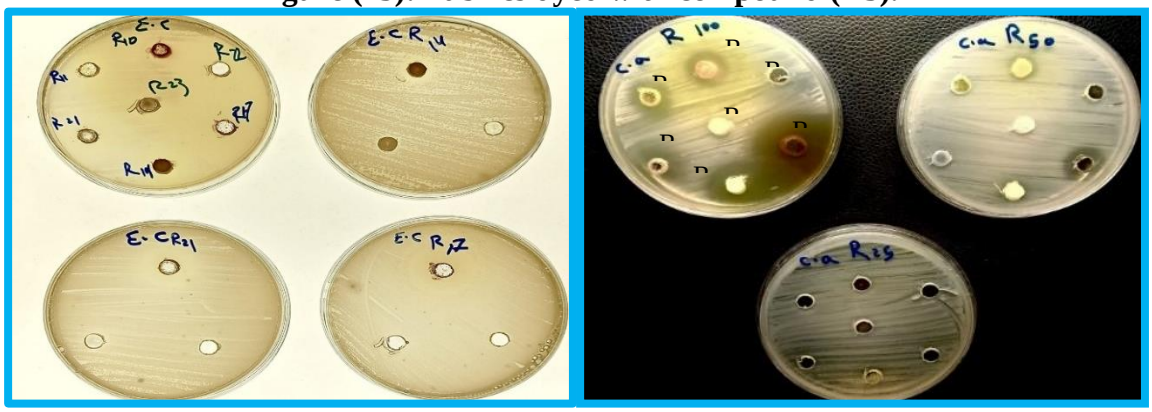


Figure (16): Inhibitory effectiveness of compounds against *E. coil* bacteria&*C. albicans.*

4. Conclusions: Most of the prepared compounds had high melting points. They were stable and consistent, as they maintained their shape, color, and melting point despite the laboratory temperature changing between the winter and summer seasons. Physical and spectroscopic measurements also confirmed the validity and accuracy of the structures. It was also found that the prepared dyes had good color stability. Reaction of derivatives of Schiff bases, and azo dyes with compounds containing suitable active groups often gives heterocyclic rings. It is also clear from the biological study that most of the prepared compounds have antibacterial activity and could inhibit bacterial growth, as it was found that some of these compounds have a higher biological effectiveness than the antibiotics used as control samples.

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