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Salicylic acid heterocyclic derivatives: chemical synthesis and biological activity

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ORIGINAL STUDY

Salicylic Acid Heterocyclic Derivatives: Chemical Synthesis and Biological Activity

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Abstract

Before a drug can be widely used, it must go through a long development process, starting from the discovery of the active ingredient and continuing with tests to confirm its safety and effectiveness. In this study, new olsalazine derivatives were synthesized, containing a fused morpholine-3-one ring and two five-membered heterocyclic rings (thiadiazole and oxadiazole). Common and readily available materials, such as salicylic acid (to prepare 2-hydroxy-5-nitrobenzoic acid) and inexpensive catalysts, were used to synthesize compounds from the first, 5,5'-(diazene-1,2-diyl) bis(2-hydroxy-3-nitrobenzoic acid) (A1), to the seventh, 6,6'-(diazene-1,2-diyl)bis(N-(4-(5-amino-1,3,4-oxadiazol-2-yl) phenyl)-3-oxo-3,4-dihydro-2H-benzo[b][1,4]oxazine-8-carboxamide) (A7).

Each compound was characterized by FT-IR and ^{1}H-NMR spectroscopy, along with physical properties such as melting point and color. All compounds showed positive results, and their antibacterial activity was tested against *Escherichia coli* and *Staphylococcus aureus*, with several demonstrating effective inhibition.

Keywords: Aromatic azo compounds, Biological activity, Olsalazine, 1,3,4-thiadiazol, 1,3,4-oxadiazol

1. Introduction

Nitrogen and sulfur have versatile frameworks for drug development and strategy [1]. Because of their aromaticity, unsaturated five-membered heterocycles are frequently more stable than other heterocyclic compounds [1]. Stability and reactivity can be influenced by the ring strain and the type of heteroatom or heteroatoms. Aromatic azo compounds are used extensively in the chemical industry as dyes, indicators, catalysts, and pharmaceuticals [2]. Moreover, azobenzenes have recently drawn attention for potential uses in the domains of molecular shuttles, chemosensors, nonlinear optics, liquid crystals, photochemical molecular switches, nanotubes, and in the manufacture of shielding eye goggles and filters [3,4]. As a share of our attention in the heterocyclic field, we aimed to prepare some newer effective derivatives of five-membered heterocyclic units in which both nitrogen atoms are bonded to one or two heterocyclic/substituted aryl groups. Unstrained 5- and 6-membered rings such as pyridine, thiophene, pyrrole, and furan are commonly used in synthetic and pharmaceutical applications [5]. The ring strain and the type of heteroatom or heteroatoms determine the compounds' stability and reactivity. Five-membered ring compounds with two heteroatoms—at least one of which is nitrogen—are known as azoles [6]. Thiazoles are defined as 5-membered rings that contain sulfur and nitrogen atoms, while oxazoles contain oxygen and nitrogen atoms in the ring. The rationale for synthesizing salicylic acid derivatives lies in the fact that it is a well-known pharmaceutical compound that can be combined with biologically active heterocyclic units to form new molecules [7–10]. The goal is to produce derivatives with improved pharmacological properties, reduced toxicity, higher efficacy, and high membrane permeability [11,12]. Despite its anti-inflammatory properties, salicylic acid can cause side effects, including gastrointestinal

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disturbances. Combining it with heterocyclic rings (such as morpholine, thiadiazole, and oxadiazole) alters the compound's polarity and molecular structure [13]. There are many modern sources used to interact salicylic acid with heterocyclic rings, focusing on the most effective methods, as in Refs. [14–17]. This modification can enhance or inhibit its efficacy, or even impart entirely new pharmacological effects [18]. The heterocyclic ring adds a new size and shape to the molecule. This new shape may better accommodate the enzyme, increasing its affinity and binding strength, and thus enhancing its efficacy [19]. The new ring may provide new sites for hydrogen bonding, hydrophobic interactions, or ionic bonding with the biological target. [20,21]. It may also increase the lipophilic properties of the lipid, facilitating its passage through cell membranes, especially if applied topically [22–24].

2. Experimental

2.1. Synthesis of samples

2.1.1. Synthesis of 2-hydroxy-5-nitrobenzoic acid

In a conical flask, 10 ml of glacial acetic acid is heated moderately in a water bath before 3 g of calcium nitrate tetrahydrate is added. After adding 2 g of salicylic acid, the reaction mixture is heated for a few minutes below 80 °C in a boiling bath of water. After producing a red solution, the dark red concoction is poured into a 100 ml beaker filled with 20 ml of ice water. After 4–5 h, yellow crystals of 4-nitrosalicylic acid separated from the opaque, dark red solution that was stored in the freezer. After filtering, the raw material is washed with cold water and allowed to dry. 4-Nitrosalicylic acid produces a yellow precipitate; yield: 75 %; melting point. 229–230 °C. The FT-IR spectrum of compound 2-hydroxy-5-nitrobenzoic acid (value, cm$'^{1}$): broad 3413-2983 (OH $_{carboxyl}$), 1737(C=$O_{carboxyl}$), 1500–1523 (N–O); ^{1}HNMR (400 MHz, DMSO-d_6) — (ppm): 5.50 (s, 1H OH phenolic), 7.80–8.85 (d, 3H, Ph), 10.50 (s, 1H, OH carboxilic).

2.1.2. Synthesis of 5-amino-2-hydroxybenzoic acid

In 200 mL of water, 33.98 g (0.185 mol) of 2-hydroxy-5-nitrobenzoic acid were dissolved with the addition of potassium hydroxide KOH until the pH reached 11.5, using Litmus paper, adding 2.0 g of ground nickel. The mixture has been refluxed with stirring and heated to 100 °C. Adding hydrazine hydrate (40 ml, 80 %) gradually (drop after drop) for a period of time, 3–4 h. After the mixture was filtered, the solution was gathered and allowed to cool to 40 °C in an ice bath.

The pH was adjusted to (2-3) by adding 35 % hydrochloric acid (20 ml), and by using Litmus paper, the solution was placed in the ice bath for 2 h to get the precipitate. Finally, after filtering the solution, the precipitate was produced, and it was purified with water and dried at 60–70 °C; Yield: 62 %; melting point 280–281 °C. The FT-IR spectrum of the compound 5-amino-2-hydroxybenzoic acid (value, cm$'^{1}$):: broad 3413–3300 ($OH_{carboxyl}$), 3315–3177 (NH_2), 1712(C=$O_{carboxyl}$); The ^{1}H-NMR spectrum for compound (400 MHz, DMSO-d_6) signals (ppm): 5.50 (s, 1H OH phenolic), 6.00 (s, 2H, NH2), 6.70–7.88 (d, 3H, Ph), 10.47 (s, 1H, OH carboxylic).

2.1.3. Synthesis of 5,5'-(diazene-1,2-diyl)bis(2-hydroxybenzoic acid) (A)

A solution of concentrated hydrochloric acid (2.25 ml) and water (4 ml) is mixed with 5.53 g (0.01 mol) of 5-amino-2-hydroxybenzoic acid. After 10 min of stirring, the resultant solution is cooled to 0–5 °C. Drop by drop, a sodium nitrite solution (0.011 mol, 0.76 g) in 2.5 ml of water is added. After 10 min of stirring, the resulting diazonium salt solution was added dropwise to a solution of salicylic acid (1.38, 0.01 mol) in ethanol and 10 % NaOH (10 ml) at 0 °C -5C – PH = 5.5. Following the addition, the mixture was agitated for an additional 20 min before being allowed to sit for an hour. The solid that resulted was then filtered and given a water wash, dried, and recrystallized from ethanol. Color: Crystalline white; Yield: 38 %; melting point. 240–241 °C; The FT-IR spectrum of compound **(A)** broad 3402–2871 ($OH_{carboxyl}$), 1711 (C=$O_{carboxyl}$), 1533–1551 (N=N stretch); ^{1}HNMR (400 MHz, DMSO-d_6) signals (ppm): 5.39 (s, 2H OH phenolic), 7.50–8.65 (d, 6H, Ph), 10.60 (s, 2H, OH carboxilic). TLC R*f* = 0.62 (n-hexane: DCM).

2.1.4. Synthesis of 5,5'-(diazene-1,2-diyl)bis(2-hydroxy-3-nitrobenzoic acid) (A1)

In a conical flask, 3 g of calcium nitrate tetrahydrate are dissolved in 10 mL of glacial acetic acid by slowly heating it in a water bath. After adding 2 g of compound A, the reaction mixture is heated for a few minutes below 80 °C in a boiling water bath. After forming a dark red solution, the mixture of chemical reactions is transferred into a 100 ml beaker filled with 20 ml of ice-cold water. After four to 5 h, the yellow crystal formed. The suction pump filters the crude product, which is then cleaned with cold water and dried. When 4-nitrosalicylic acid is recrystallized from methanol, a yellow precipitate is produced. Color: white crystal; Yield. After four to 5 h, the yellow crystals of 4-nitrosalicylic acid separated

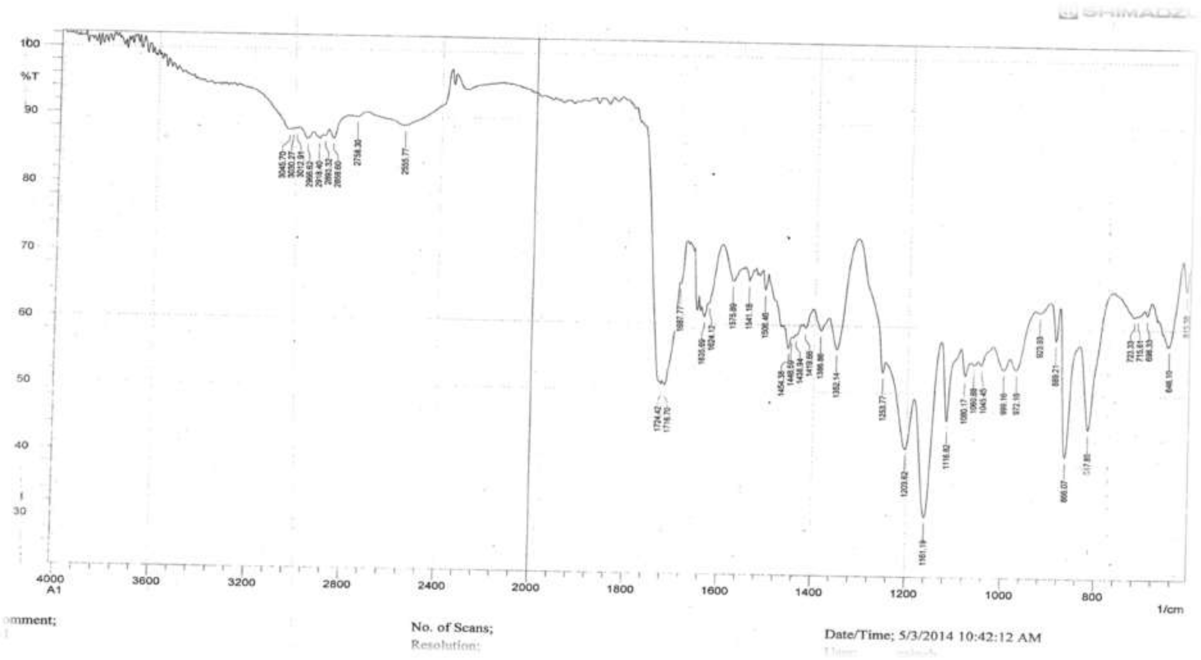

Fig. 1. FT-IR spectrum of compound A1.

from the turbid, dark red solution that had formed. Color: white crystal; Yield: 51 %; melting point. 213–214 °C; The FT-IR spectrum of compound A1 (Fig. 1) (value, cm′ 1): broad 3388–2911 ($OH_{carboxyl}$), 1677($C{=}O_{carboxyl}$), 1568–1560 (N=N stretch); ^{1}HNMR (Fig. 5) (400 MHz, DMSO-d_6) signals (ppm):5.67 (s, 2H OH phenolic), 8.50–8.85 (d, 4H, Ph), 10.22 (s, 2H, OH carboxylic).

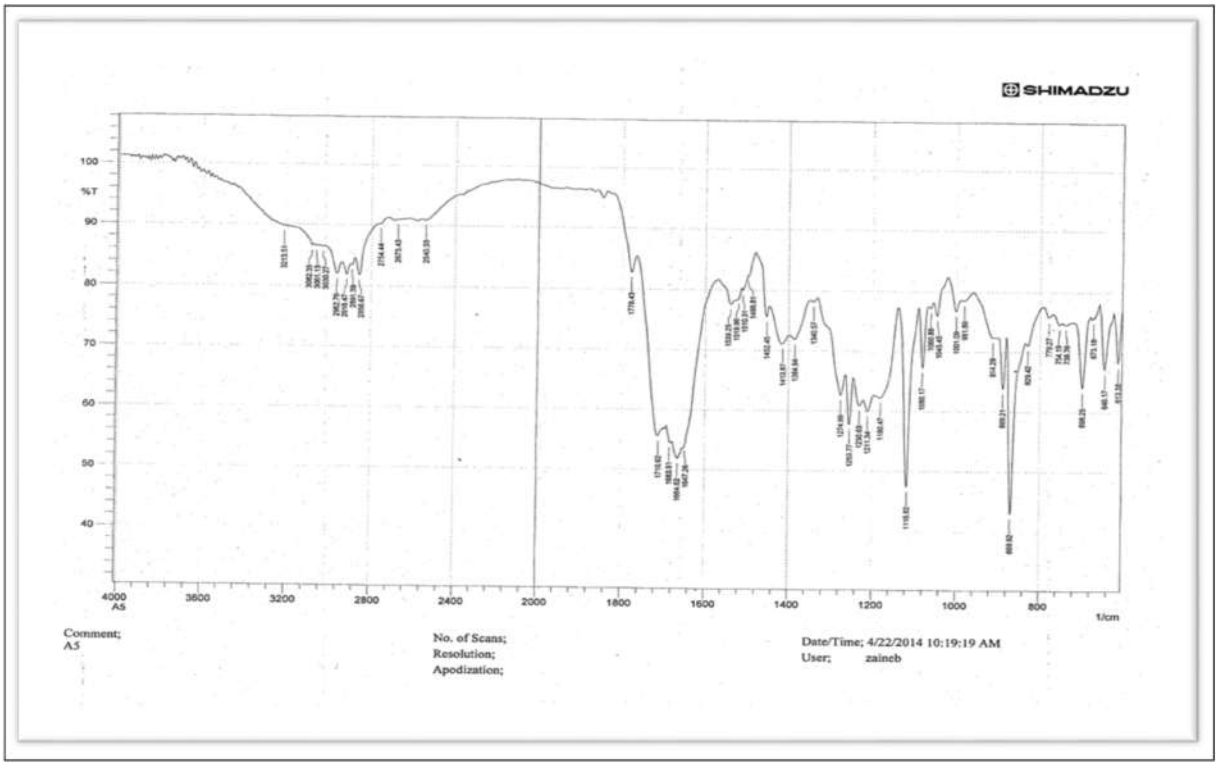

Fig. 2. FT-IR spectrum of compound A3.

2.1.5. Synthesis of 5,5'-(diazene-1,2-diyl)bis(3-amino-2-hydroxybenzoic acid) (A2)

Compound A1 (3.92 g, 0.01 mol) was dissolved in 20 ml) of water with the addition of KOH until the pH reached 1.15, using Litmus paper, with the addition (0.2 g of ground nickel. The mixture has been refluxed with stirring and heated to 100 °C. Adding hydrazine hydrate (4 ml, 80 %) gradually for 3–4 h. The combination was filtered, and the solution was collected and placed in an ice bath at 40 °C.

The pH was adjusted to (2-3) by adding 35 % hydrochloric acid (2 ml), and by using Litmus paper, the solution was placed in an ice bath for 2 h to get the precipitate. Finally, the solution was filtered, the precipitate was obtained, and it was purified with water and dried at 60–70 °C. Yield: 38 %; melting point. 183–184 °C; The FT-IR spectrum of compound (A2) (value, cm$'^{1}$): broad 3411–3311 ($OH_{carboxyl}$), 3313–3178 (NH_2), 1720 ($C{=}O_{carboxyl}$) 1570–1590 (N═N stretch); ^{1}HNMR (400 MHz, DMSO-d_6) signals (ppm):5.60 (s, 2H OH phenolic), 6.00 (s, 4H, NH_2), 7.61–7.75 (d, 4H, Ph), 10.88 (s, 2H, OH carboxylic).

2.1.6. Synthesis of 5,5'-(diazene-1,2-diyl)bis(3-(2-chloroacetamido)-2-hydroxybenzoic acid) (A3)

In a conical flask, 3 g of calcium nitrate tetrahydrate are dissolved in 10 mL of glacial acetic acid by slowly heating it in a water bath. After adding 2 g of compound A, the reaction mixture is heated for a few minutes below 80 °C in a boiling water bath. After forming a dark red solution, the reaction mixture is transferred into a 100 ml beaker filled with 20 ml of ice-cold water. After four to 5 h, the yellow crystal formed. The suction pump filters the crude product, which is then cleaned with cold water and dried. When 4-nitrosalicylic acid is recrystallized from methanol, a yellow precipitate is produced. Color: white crystal; Yield. After four to 5 h, the yellow crystals of 4-nitrosalicylic acid separated from the turbid, dark red solution that had formed. Color: white crystal; Yield: 46 %; melting point. 198–199 °C; The FT-IR spectrum of compound A3 (Fig. 2) (value, cm$'^{1}$):: broad 3450–3320 ($OH_{carboxyl}$), 1688($C{=}O_{carboxyl}$), 1629 (C═Oamid), 1548–1548 (N═N stretch); ^{1}HNMR (Fig. 6) (400 MHz, DMSO-d_6) signals (ppm):4.5 (s, 4H CH_2Cl) 5.90 (s, 2H OH phenolic), 7.03 (s, 2H, NH), 8.60–8.94 (d, 4H, Ph), 10.33 (s, 2H, OH carboxylic).

2.1.7. Synthesis of 6,6'-(diazene-1,2-diyl)bis(3-oxo-3,4-dihydro-2H-benzo[b][1,4]oxazine-8-carboxylic acid) (A4)

After adding Compound A3 (0.007, 3.63 g) to a mixture of DMF (35 ml) and K_2CO_3 (2.1 g, 0.015 mol)

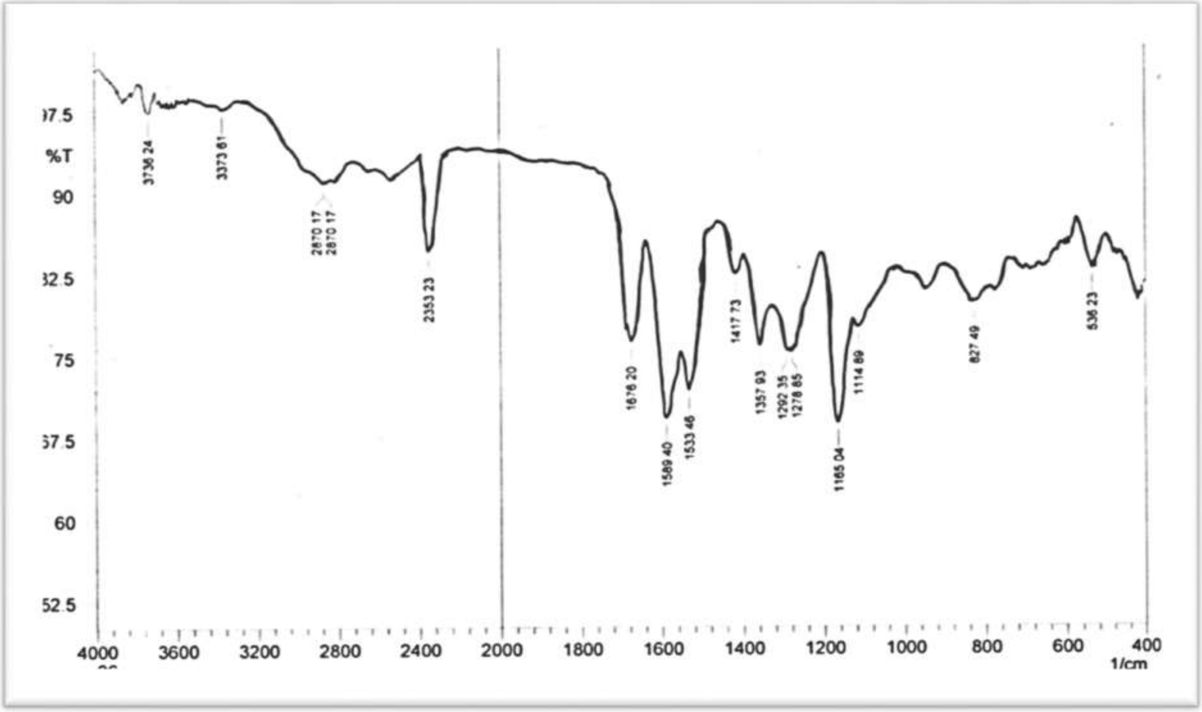

Fig. 3. FT-IR spectrum of compound A5.

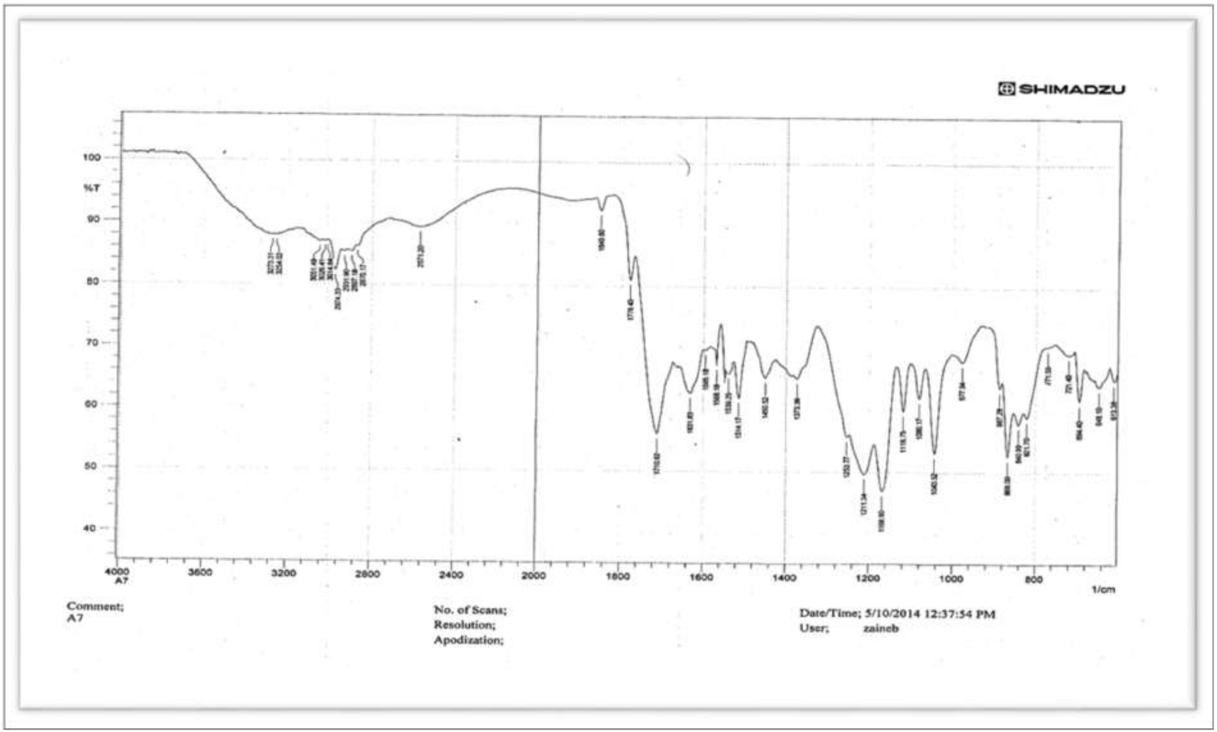

Fig. 4. FT-IR spectrum of compound A7.

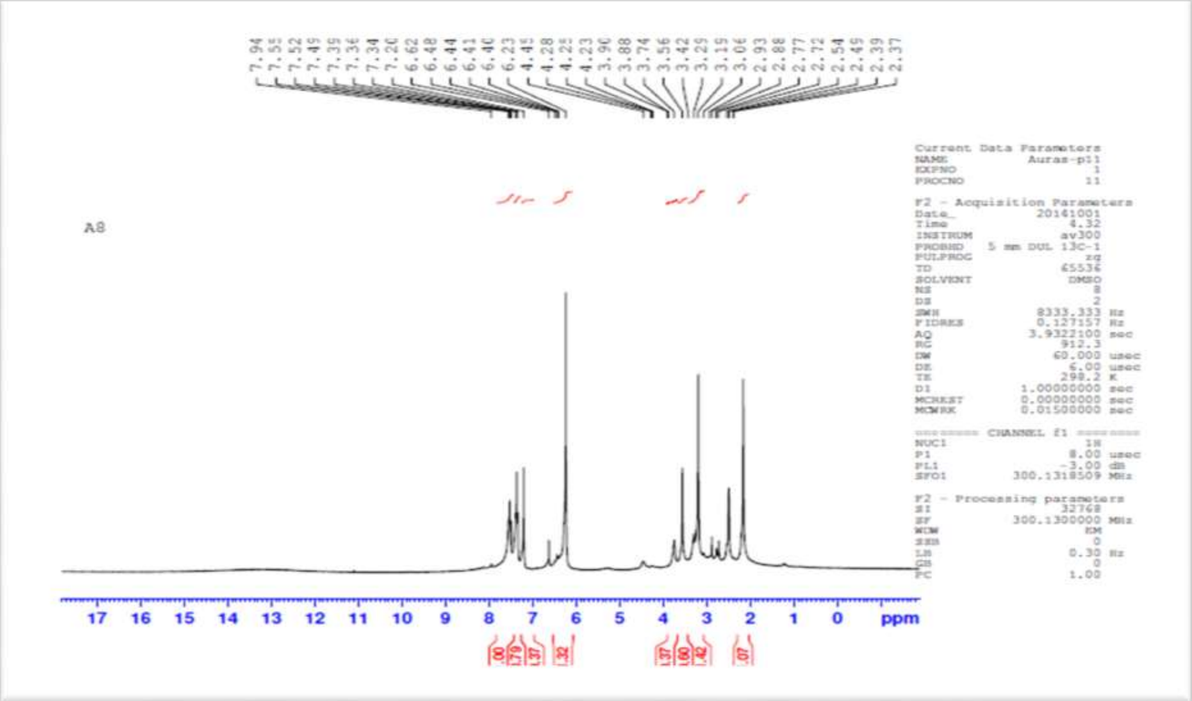

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Fig. 5. ^{1}H-NMR spectrum of compound A1.

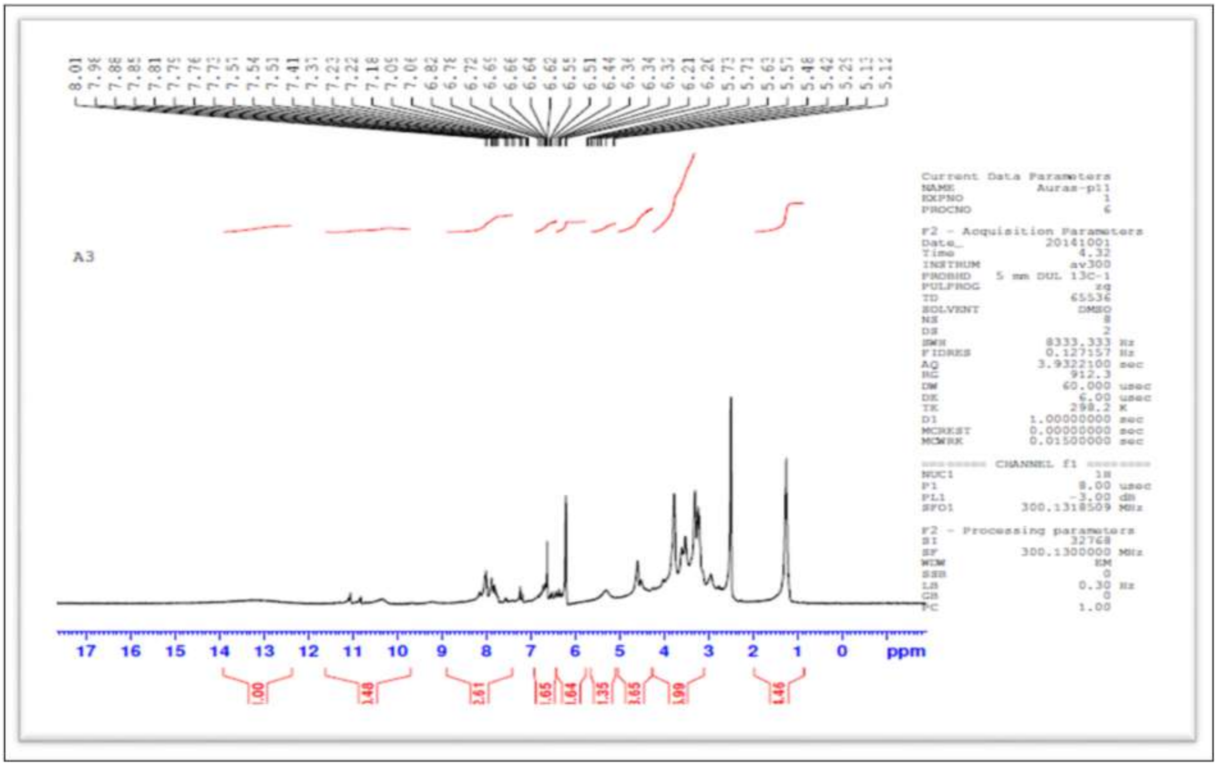

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Fig. 6. ^{1}H-NMR spectrum of compound A3.

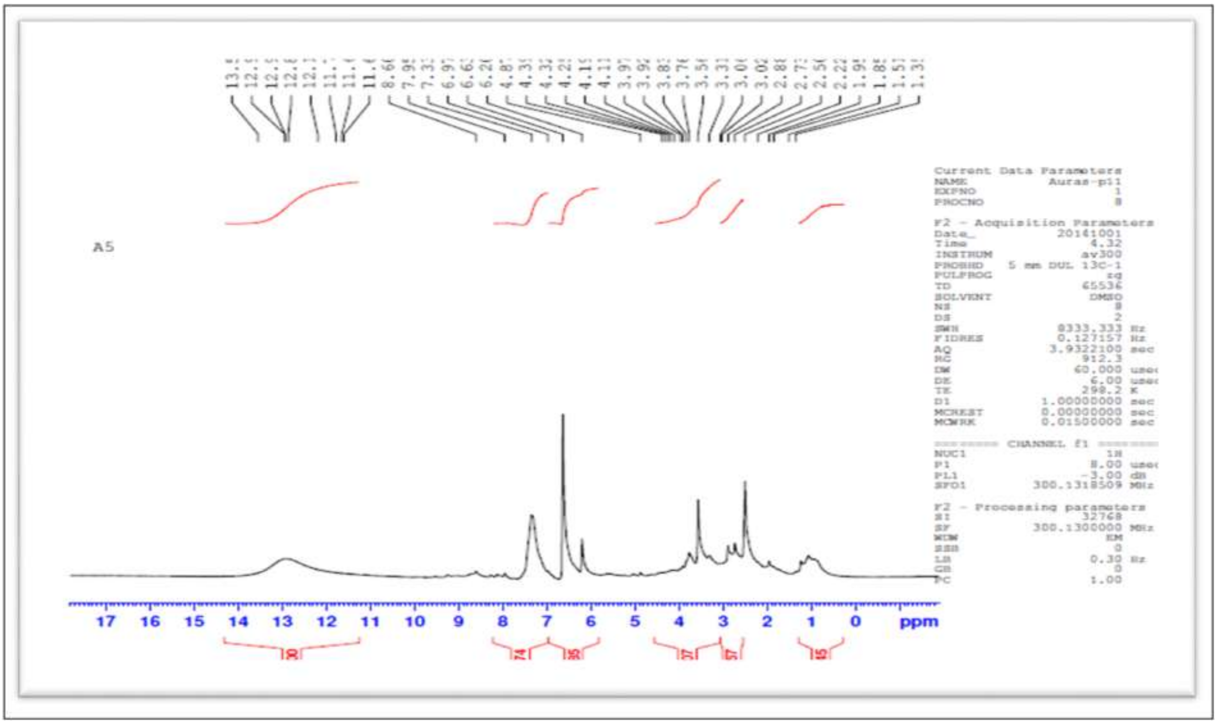

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Fig. 7. ^{1}H-NMR spectrum of compound A5.

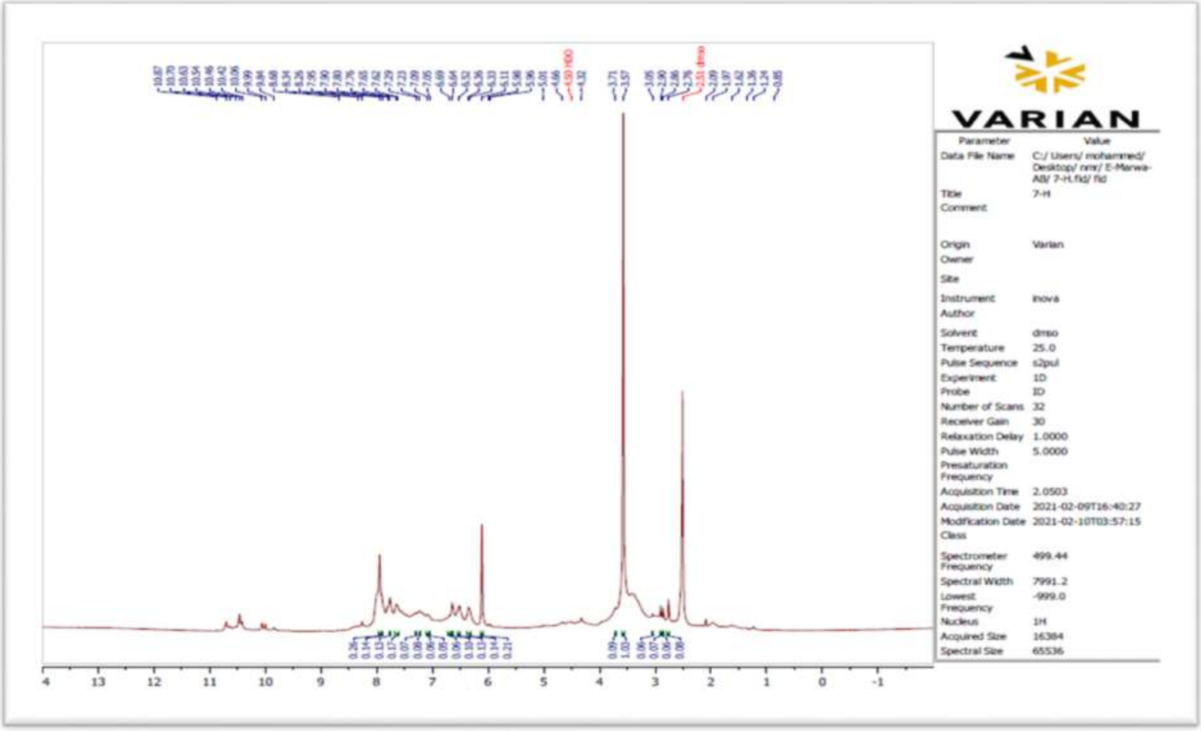

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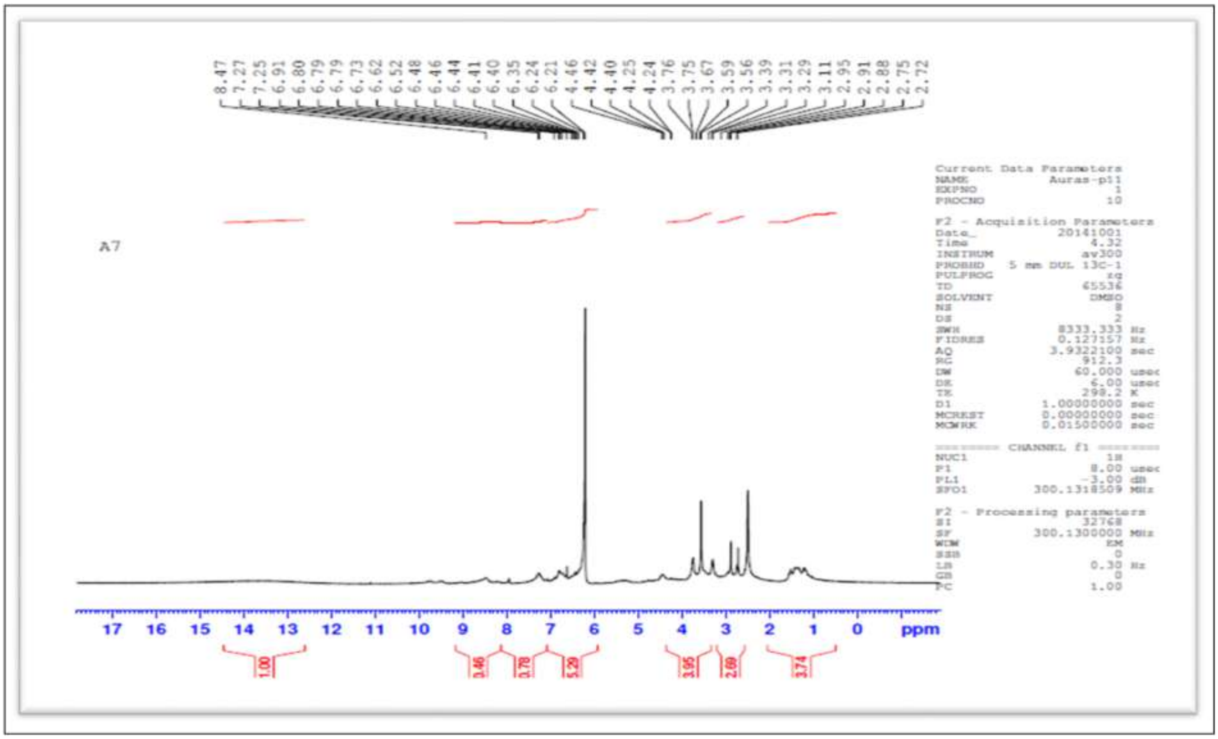
Fig. 8. ^{1}H-NMR spectrum of compound A6.



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PC                     1.00
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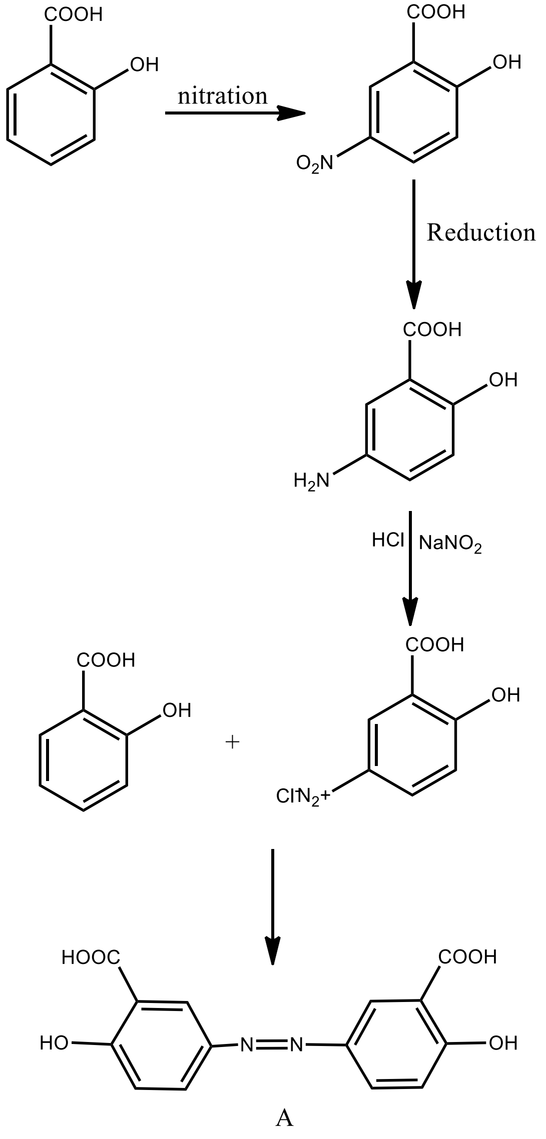
Fig. 9. ^{1}H-NMR spectrum of compound A7.

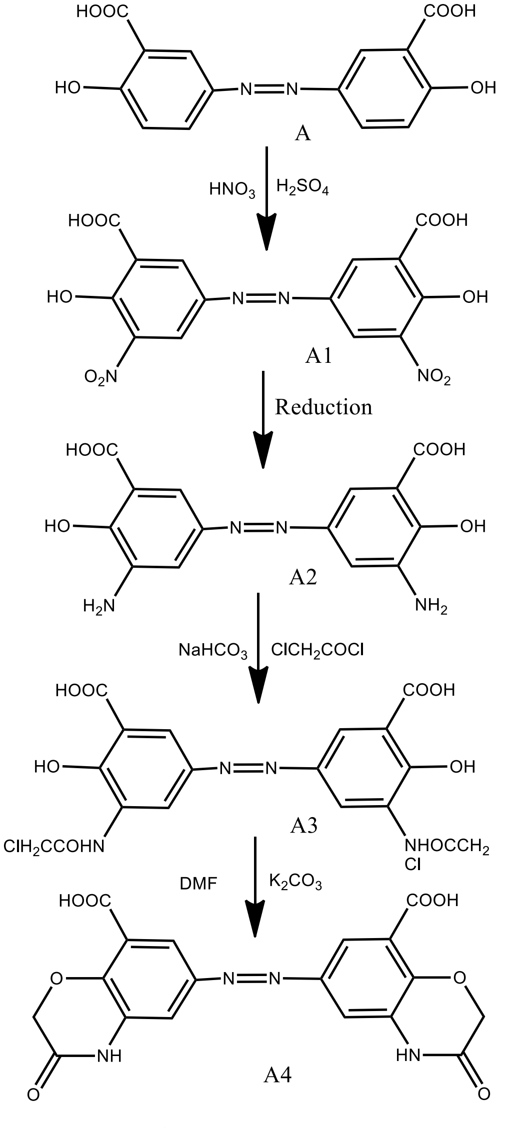
while stirring at room temperature, the mixture was heated for three more hours at 70–75 °C. A two-layer mixture of 50 ml of ice water and 50 ml of AcOEt was created by decanting the mixture. After separating the organic layer, it was cleaned with water and saturated brine in turn before being dried on top of $MgSO_4$. To obtain a solid that was recrystallized from ethanol-IPE, the organic layer was vacuum-evaporated. Produce: 37 %; melting point. 190–192 °C; The FT-IR spectrum of compound (A4) (value, cm$'^{1}$) broad 3400–3309 ($OH_{carboxyl}$), 3382 (NH) 1722($C{=}O_{carboxyl}$), 1555–1522 (N=N stretch); ^{1}HNMR (400 MHz, DMSO-d_6) signals (ppm):4.8 (s, 4H CH_2 oxazine) 7.90 (s, 2H, NH), 8.10–8.35 (d, 4H, Ph), 10.50 (s, 2H, OH carboxylic).

2.1.8. Synthesis of diethyl 4,4′-((6,6′-(diazene-1,2-diyl)bis(3-oxo-3,4-dihydro-2H-benzo[b][1,4]oxazine-8,8 -carbonyl))bis(azanediyl))dibenzoate (A5)

After dissolving (0.005 mol, 2.00 g) of compound A4 in 35 ml of thionyl chloride in a 50 ml round-bottom flask, the mixture was refluxed for 3 h before the solution was rotary-evaporated, and the oil product was collected. Drop by drop, dissolve 0.01 mol (1.65 g) of ethyl 4-aminobenzoate in 25 ml of dioxane over 30 min while stirring for 24 h. After that, it was added to the ice water and stirred for 30 min before being acidified with strong hydrochloric acid. The TLC technique was used to monitor the reaction. Diethyl ether was used to extract the organic layer, and the extraction process was repeated four times. Production: 68 %; milting



Scheme 1. Synthesis of compound (A) from salicylic acid.

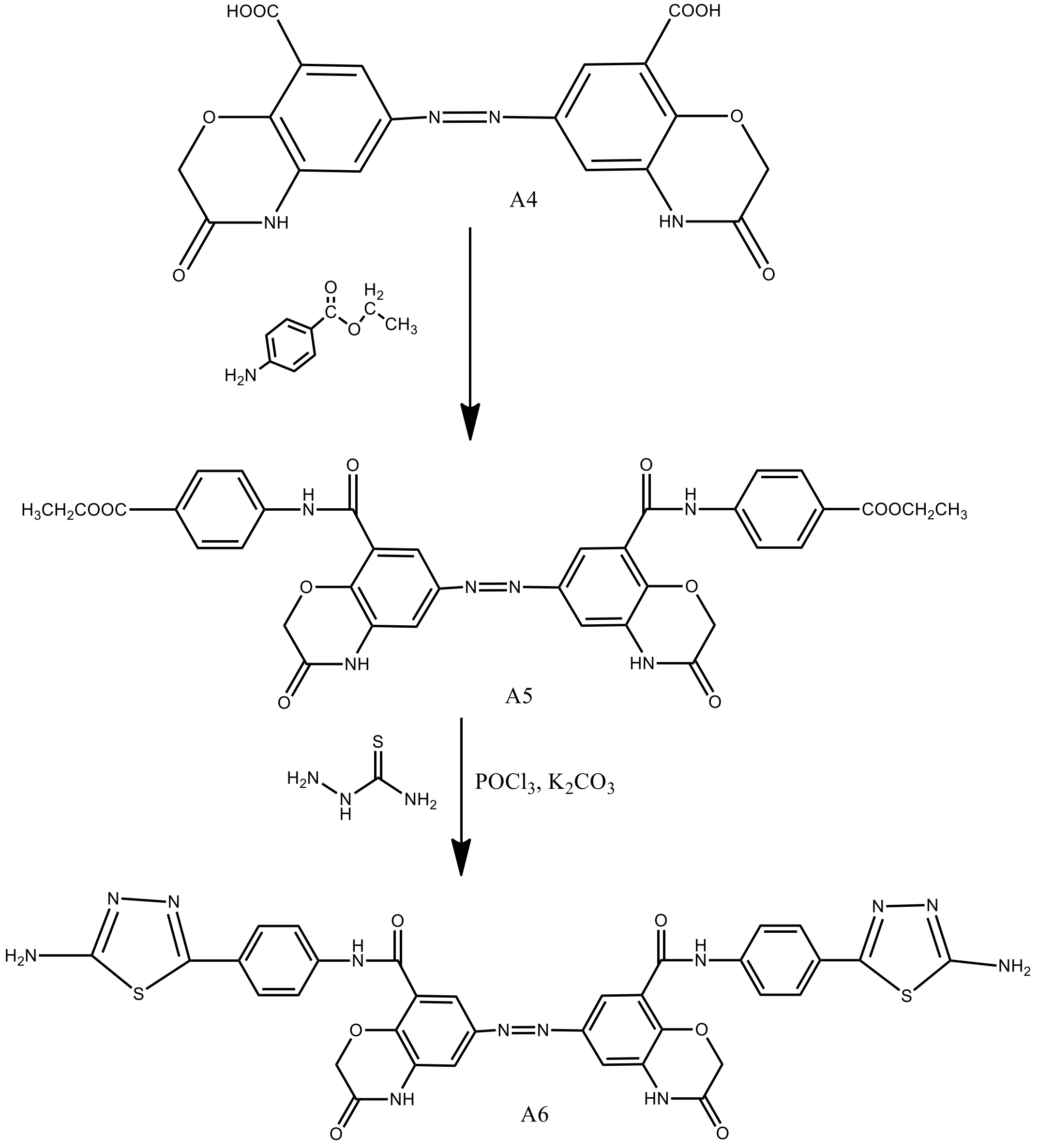


Scheme 2. Preparation of A1 -A4 via nitration, reduction, and acetylation of compound (A).

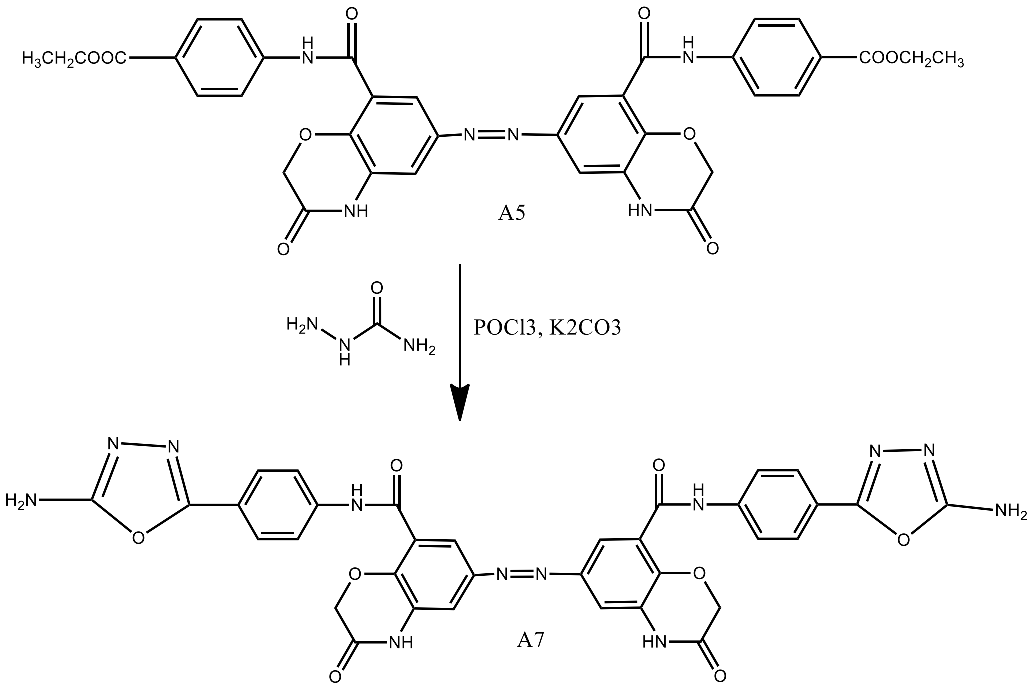
point 223–224 °C; The FT-IR spectrum of compound A5 (Fig. 3) (value, cm′ ^{1}): 3396 (NH), 2983–2687 (C–H aliph.), 3088 (C-Har), 1652 (C=O amid), 1544–1560 (N=N stretch); ^{1}HNMR (Fig. 7) (400 MHz, DMSO-d_6) signals (ppm): 1.97 (t, 6H, CH3), 4.51(m, 6H,/ CH$_2$/ CO), 4.8 (s, 4H, CH$_2$ oxazine) 7.90 (s, 2H, NH oxazine), 8.10–8.35 (d, 12H, Ph), 9.50 (s, 2H, NH imide).

2.1.9. Synthesis of 6,6′-(diazene-1,2-diyl)bis(N-(4-(5-amino-1,3,4-thiadiazol-2-yl)phenyl)-3-oxo-3,4-dihydro-2H-benzo[b][1,4]oxazine-8-carboxamide) (A6)

Compound A5 (0.005 mol, 3.53 g) and thiosemicarbazide (0.01 mol, 0.91 g) were dissolved in 15 ml of POCl$_3$, and the mixture was refluxed for 36 h. After slowly pouring the mixture onto crushed ice while stirring constantly, potassium carbonate (K_2CO_3) was added to neutralize the acid mixture. The mixture was then allowed to settle for 12 h before being filtered to extract the precipitate. Following a distilled water wash, the precipitate was recrystallized from ethanol. Production: 60 %; melting point. 166–168 °C; The FT-IR spectrum of compound (A6) (value, cm′ ^{1})3316–3166 (NH2), 3066 (C-Har), 1644 (C=N), 1577–1541 (N=N stretch); ^{1}HNMR (Fig. 8) (400 MHz, DMSO-d_6) signals (ppm): 4.8 (s, 4H, CH$_2$ oxazine), 6.5 (s, 4H, NH$_2$-thiadiazol), 7.90 (s, 2H, NH oxazine), 8.11–8.55 (d, 12H, Ph), 9.30 (s, 2H, NH imide).



Scheme 3. Synthesis of compounds (A5) and (A6) from compound (A4).



Scheme 4. Synthesis of compound (A7) from compound (A5).

2.1.10. Synthesis of 6,6'-(diazene-1,2-diyl)bis(N-(4-(5-amino-1,3,4-oxadiazol-2-yl)phenyl)-3-oxo-3,4-dihydro-2H-benzo[b][1,4]oxazine-8-carboxamide) (A7)

Compound A5 (0.005 mol, 3.53 g) and semicarbazide (0.01 mol, 0.71 g) were dissolved in 15 ml of POCl3, and the mixture was refluxed for 36 h. After slowly pouring the mixture onto crushed ice while stirring constantly, potassium carbonate (K_2CO_3) was added to neutralize the acid mixture. The mixture was then allowed to settle for 12 h before being filtered to extract the precipitate. Following a distilled water wash, the precipitate was recrystallized from ethanol. Production: 32 %; melting point. 197–199 °C The FT-IR spectrum of compound A7 (Fig. 4) (value, cm′ ^{1}): 3304–3187 (NH_2), 3029 (C-Har), 1666 (C=N), 1544–1542 (N=N stretch); ^{1}HNMR (Fig. 9) (400 MHz, DMSO-d_6) signals (ppm):4.52 (s, 4H, CH_2 oxazine), 6.63 (s, 4H, NH_2-oxadiazol), 7.84 (s, 2H, NH oxazine), 8.20–8.55 (d, 12H, Ph), 9.77 (s, 2H, NH imide).

3. Results and discussion

3.1. 1FTIR and ^{1}H-NMR characterizations

Schemes 1–4 were followed in the production of the nominated compounds.

In particular, the impact and effectiveness of bacteriophage antibodies against *Staphylococcus aureus* and *Escherichia coli* were examined. As indicated in Table 1, a number of these compounds exhibited antagonistic effects (see Table 2).

These types of bacteria were selected, one of which is Gram-positive (*Staphylococcus aureus*) and

Table 1. Shows the inhibition of the growth of the bacteria (Inhibition Zone) by some derivatives recorded in millimeter units.

Comp. No.	Type of bacteria	
	Escherichia coli	*Staphylococcus aureus*
Cefotaxime (Antibiotic) Standard	0	0
A	18	18
A1	0	0
A2	10	20
A3	0	25
A4	15	12
A5	30	25
A6	15	18
A7	20	20

Table 2. Shows the inhibition of the growth of the fungal (Inhibition Zone) by some derivatives recorded in millimeters.

Comp. No.	Fungal Aspergillus Niger
A	15
A1	18
A2	20
A3	20
A4	10
A5	10
A6	15
A7	15

the other Gram-negative (*Escherichia coli*), which cause several infections.

The degree of inhibition of bacterial growth was measured by the products synthesized and related to cefotaxime. The agar diffusion method was used to evaluate the biological effectiveness of the synthesized products. Most of the synthesized compounds were found to have biological effects as inhibitors of the growth of these two types of bacteria. When compared to cefotaxime's biological effectiveness, it was discovered that the synthesized compounds A5 and A7 had a high zone of inhibition for *Escherichia coli*. Additionally, it was discovered that, in comparison to cefotaxime, the synthesized compounds A1, A4, and A6 are highly effective at inhibiting the growth of *Staphylococcus aureus*. Furthermore, most of the prepared compounds were found to have biological effects as inhibitors for the growth of the fungus Aspergillus niger.

4. Conclusions

Various products made from salicylic acid could be used to prepare other compounds because they were suitable intermediates, and the yield of products was sufficient for the next stage. The products prepared under ambient conditions are not affected by moisture or light, and the high melting points of some products indicate their stability. Selected biologically active synthesized compounds can be used because they have a high ability to inhibit the growth of *Escherichia coli* and *Staphylococcus aureus*

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Conflict of Interest

The authors declare that there is no conflict of interest regarding this research.

Ethical Approval

The study was conducted in accordance with ethical standards. This research does not involve experiments on humans or animals.

Data Availability

All data used in this study are included within the manuscript and can be obtained from the authors upon request.

Author Contributions

All authors contributed to the preparation of the manuscript, conducting the practical work, analyzing the results, and writing the final review of the study.

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References

[1] Mallappa, Chahar M, Choudhary N, Yadav KK, Qasim MT, Zairov R, et al. Recent advances in the synthesis of nitrogen-containing heterocyclic compounds via multicomponent reaction and their emerging biological applications: a review. J Iran Chem Soc 2025;22(1):1–33. https://doi.org/10.1007/s13738-024-03142-3.

[2] De Joarder D, Sarkar R, Maiti DK. Synthesis of bioactive heterocycles involving heterogeneous catalysis in water. Aqueous-Mediated Synth Bioact Heterocycles 2024;2(1):307. https://doi.org/10.1039/x0xx00000x.

[3] Duc DX, Quoc N Van. Isolation, bioactivities, and synthesis of lamellarin alkaloids: a review. Curr Org Chem 2022;26(10): 961–90. https://doi.org/10.2174/1385272826666220516114926.

[4] Gupta RR, Kumar M, Gupta V. Heterocyclic chemistry: volume II: five-membered heterocycles. Springer Science & Business Media; 2013.

[5] Ali Y, Hamid SA, Rashid U. Biomedical applications of aromatic azo compounds. Mini Rev Med Chem 2018;18(18):1548–58. https://doi.org/10.2174/1385272826666220516114926.

[6] Li Petri G, Raimondi MV, Spano V, Holl R, Barraja P, Montalbano A. Pyrrolidine in drug discovery: a versatile scaffold for novel biologically active compounds. Top Curr Chem 2021;379(5):34. https://doi.org/10.1007/s41061-021-00347-5.

[7] Giles RL, Nkansah RA, Looper RE. Synthesis of 2-Thio-and 2-Oxoimidazoles via Cascade addition/ cycloisomerization reactions of propargylcyanamides. J Org Chem 2010;75(1): 261–4. https://doi.org/10.1021/jo902326d.

[8] Rasool SR, Aljamali NM, Al-Zuhairi AJ. Guanine substituted heterocyclic derivatives as bioactive compounds. Biochem Cell Arch 2020;20(Supplement 2):3651–5. https://doi.org/10.1021/jo902326d.

[9] Naz S, Shah FA, Nadeem H, Sarwar S, Tan Z, Imran M, et al. Amino acid conjugates of aminothiazole and aminopyridine as potential anticancer agents: synthesis, molecular docking and in vitro evaluation. Drug Des Devel Ther 2021:1459–76. https://doi.org/10.2147/DDDT.S297013.

[10] Cortez-Maya S, Pedro-Hernandez LD, Martínez-Klimova E, Ramírez-Apan T, Martínez-García M. Anticancer activity of water-soluble olsalazine-PAMAM-dendrimer-salicylic acid-conjugates. Biomolecules 2019;9(8):360. https://doi.org/10.3390/biom9080360.

[11] Sanad MH, Marzook FAM, Challan SB, Essam HM, Farag AB. Radioiodination, and biological assessment of olsalazine, as a highly selective radiotracer for ulcerative colitis imaging in mice. Arab J Nucl Sci Appl 2023;56(3): 105–20. https://doi.org/10.21608/ajnsa.2022.163538.1639.

[12] Romero-Castro A, Gutierrez-Sanchez M, Correa-Basurto J, Rosales Hernandez MC, Padilla Martínez II, Mendieta-Wejebe JE. Pharmacokinetics in wistar rats of 5-[(4-carboxybutanoyl) amino]-2-hydroxybenzoic acid: a novel synthetic derivative of 5-aminosalicylic acid (5-ASA) with possible anti-inflammatory activity. PLoS One 2016;11(7):e0159889. https://doi.org/10.1371/journal.pone.0159889.

[13] V N, Fedoseeva VG, Verochkina EA. The reactions of p-Tosylmethyl isocyanide with aldehydes in the synthesis of heterocyclic compounds: a review. Mini Rev Org Chem 2023;20(4):372–93. https://doi.org/10.2174/1570193X19999220523113243.

[14] Mishra AK, Baek KH. Salicylic acid biosynthesis and metabolism: a divergent pathway for plants and bacteria. Biomolecules 2021;11(5). https://doi.org/10.3390/biom11050705.

[15] https://pubs.acs.org/doi/abs/10.1021/acs.orglett.2c02066#:~:text=4*Done%20Derivatives-, Masahiro%20Abe, Kiyofumi %20Inamoto *. Gold(I)-Catalyzed Heteroannulation of Salicylic Amides with Alkynes: Synthesis of 1,3-Benzoxazin-4-one Derivatives. . doi:10.1021/acs.orglett.2c02066.

[16] Abe M, Kawamoto M, Inoue M, Kimachi T, Inamoto K. Gold (I)-Catalyzed heteroannulation of salicylic amides with alkynes: synthesis of 1,3-Benzoxazin-4-one derivatives. Org Lett [Internet] 2022 Aug 12;24(31):5684–7. https://doi.org/10.1021/acs.orglett.2c02066.

[17] Muheyuddeen G, Khan MY, Ahmad T, Srivastava S, Verma S, Ansari MS, et al. Design, synthesis, and biological evaluation of novel imidazole derivatives as analgesic and anti-inflammatory agents: experimental and molecular docking insights. Sci Rep [Internet] 2024;14(1):23121. https://doi.org/10.1038/s41598-024-72399-8.

[18] Chugh V, Pandey G, Rautela R, Mohan C. Heterocyclic compounds containing thiazole ring as important material in medicinal chemistry. Mater Today Proc 2022;69:478–81. https://doi.org/10.1016/j.matpr.2022.09.150.

[19] Ahmed SM, Hasan HM. Archive of SID. ir. 2023. https://doi.org/10.26655/JMCHEMSCI.2023.7.6.

[20] AL-Abayechi MMH, AL-Zuhairi AJ, AL-Baiati MN. Synthesis and identification of a novel hyper branched polymers containing melamine derivative. In: IOP conference series: materials science and engineering. IOP Publishing; 2019. p. 12092. https://doi.org/10.1088/1757-899X/571/1/012092.

[21] Pierozan P, Colín-Gonzalez AL, Biasibetti H, da Silva JC, Wyse A, Wajner M, et al. Toxic synergism between quinolinic acid and glutaric acid in neuronal cells is mediated by oxidative stress: insights to a new toxic model. Mol Neurobiol 2018;55(6):5362–76. https://doi.org/10.1007/s12035-017-0761-6.

[22] dos Santos JM, Visentin APV, Scariot FJ, Echeverrigaray S, Salvador M, Branco CS. The effect of different polyphenols against neurotoxicity induced by quinolinic acid in U87-MG glial cells. Res Soc Dev 2022;11(1). https://doi.org/10.33448/rsd-v11i1.24865.

[23] Orozco-Morales M, Hernandez-Pedro NY, Barrios-Bernal P, Arrieta O, Ruiz-Godoy LM, Aschner M, et al. S-allylcysteine induces cytotoxic effects in two human lung cancer cell lines via induction of oxidative damage, downregulation of Nrf2 and NF- B, and apoptosis. Anti Cancer Drugs 2021;32(2): 117–26. https://doi.org/10.1097/CAD.0000000000001015.

[24] Ali Y, Hamid SA, Rashid U. Biomedical applications of aromatic azo compounds. Mini Rev Med Chem 2018;18 (18):1548–58. https://doi.org/10.2174/138955751866618052411 3111.