Synthesis, Characterization, Novel Oxazolone Derivatives of Study Cytotoxicity, Antioxidant and Antibacterial Activity in Vitro

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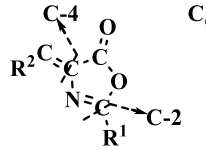
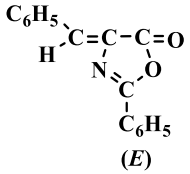
Abstract.
By reaction aromatic aldehydes with glycine in the presence of acetic anhydride and anhydrous sodium acetate, a new family of Mono-Bis oxazolone molecules was successfully produced. The spectrum data supported the synthesis of these compounds, and the melting point, IR, 1H-NMR,13C-NMR & Mass were used to identify this Mono-Bis oxazolone chemical. Hemolysis study demonstrated that the hemolysis percentage of Mono-Bis oxazolone at (0.5 mg/ml) all concentrations were less than 4%, this result indicates the safety of their use inside the body. The antioxidant activity of the Mono-Bis oxazolone compounds against DPPH radicals was tested in vitro. The results a good.
Keywords: oxazolones, hippuric acid, fused sodium acetate, Mono-Bis, Antioxidant & Cytotoxicity and Vitro.

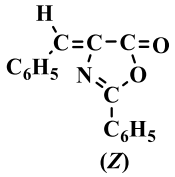
1. Introduction

Oxazolones are a type of tiny heterocyclic compounds that have gained increased attention in recent years due to their pharmacological actions. Oxazolones are mostly Nitrogen and Oxygen containing five & six-member heterocyclic compounds[1]. It belongs to a broad family of oxazole-based chemicals and was called according to Hantzsch-Widman nomenclature [2]. of physiologically active chemicals. It has a wide range of pharmacological properties, including antibacterial, antifungal, anti-diabetic, anti-cancer, and anti-inflammatory [3]. The biological effects of oxazolone are attributed to the C_{-2} and C_{-4} positions, which include analgesic [4], anti-inflammatory, antidepressant [5], anticancer, antimicrobial [6], antidiabetic [7], and antiobesity. Unsaturated Oxazalone exist as (*E*) and (*Z*) geometric isomers in compounds [8, 9], respectively, where they have been isolated. History of oxazalone. Plo chl produced oxazolone for the first time in 1883 by condensation of benzaldehyde with hippuric acid in the presence of acetic anhydride19,20. Friedrich Gustav Carl Emil Erlenmeyer established the first correct structure of oxazolone in 1893 by reacting benzaldehyde with N-acetylglycine in the presence of acetic anhydride and sodium acetate, followed by Perkins condensation and initial cyclisation of N-acetylglycine, yielding Erlenmeyer oxazolone [10].



Structure of Oxazalone C-2 and C-4 positions,





E- & Z- isomers of unsaturated oxazolone

2. MATERIALS AND METHODS.

2.1. Instrumentation:

Merck (Germany), Sigma-Aldrich (USA), and BDH provided all of the materials (UK). The melting point was measured using the melting point equipment. FT-IR spectra were obtained using potassium bromide (KBr) discs (4000–400) cm-1 on a Shimadzu FT-IR (8300) spectrophotometer. ^{13}CNMR and ^{1}HNMR spectra were acquired using $CDCl_3$ and DMSO-d_6 as solvents, Tetramethylsilane (TMS) as an internal reference, and chemical shift was quantified in ppm relative to TMS on a Bruker(400MHz) NMR spectrophotometer. The GC-Mass spectra were obtained at 70 eV with an Agilent technologies mass selective detector 5973 wett work at Technology Shareef University in Tehran, Iran.

2.2: Procedures:

2.2.1: General Procedures for the preparations of Oxazolone13.

2.2.1.1: Preparation method of Mono-Bis oxazolone.

In general, mono-bis oxazolone is made by, 20 ml of (Ethanol+ Methanol), an equimolecular mixture of glycine (0.01 or 0.02mol) derivatives aromatic aldehyde (0.01 or 0.02mol) and derivatives benzoyl chloride(0.01mol) was refluxed with constant stirring for (4-12) hrs.' And sodium acetate (0.01 or 0.02mol). In the presence of a few drops of acetic anhydride. TLC is used to monitor the reaction's progress. The eluent is hexane:ethyl acetate 3:7, which evaporates and is then recrystallized from a suitable solvent, The oxazolone structures were validated by comparing their m.p, mixed. m.p, TLC, IR, ^{1}H-NMR & ^{13}C-NMR data to those in the literature. The physical data of mono-bis oxazolone B(1-6) as shown in Table (1). The following methods of mono-bis oxazolone preparations are:

2.2.1.1.1: 4-(4-(Dimethylamino)benzylidene)-2-(4-hydroxyphenyl)oxazol-5(4H)-one(1B). It reactions were used to create. A few drops of acetic anhydride, glycine(0.01mol, 0.75g), 4-(dimethylamino)benzaldehyde(0.01mol, 1.49g), 4-

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hydroxy benzoylchloride(0.01mol, 1.56g) and fused sodium acetate(0.01mol, 0.8g). m.p.= 258-260^{0}C. R_f = 0.7. Yield= 85%. IR (ῡ, cm ^{1}, KBr disk). 3383(OH stretching vibration), 3043,3020(sp^2-CH stretching frequency), 2965,2899(sp^3-CH stretching frequency), 1748(C=O), 1669(C=N), 1599(C=C). ^{1}H-NMR (400MHz, DMSO-d_6): δ(*s*, 3.03ppm, 6H, H_3C-N-CH_3), δ(*dd*, 6.76-6.78ppm, J=8, 2H) Para Para System, δ(*dd,*7.66-7.68ppm, J=8, 2H+1H), Para Para System+C=CH), δ(*dd*, 8.15-8.17ppm, J=8, 2H) Para Para System δ(*dd*, 8.30-8.32ppm, J=8, 2H), Para Para System, δ(*s*, 9.65ppm, 1H, OH).C-NMR (400MHz, DMSO-d_6): δ(39.76 ppm, C-N-C) δ(110.69-136.76 ppm, aromatic proton), δ(166.01 ppm, C=O), δ(164.34 ppm, C=N), δ(150.10 ppm, C=C).

2.2.1: 4-(Cyclopentylmethylene)-2-(4-nitrophenyl) oxazol-5(4*H*)-one(2B).

It reactions were used to create. A few drops of acetic anhydride, glycine (0.01mol, 0.75g), cyclopentanecarbaldehyde(0.01mol, 0.98g, 1.06ml), 4-nitrobenzoyl chloride(0.01mol, 1.85g) and sodium acetate(0.01mol, 0.8g). m.p.= 93-95^{0}C. R_f = 0.9. Yield= 82%. IR (ῡ, cm^{-1}, KBr disk), 3079,3115(sp^2-CH stretching frequency), 2840,2993(sp^3-CH stretching frequency), 1695(C=O), 1606(C=N), 1541(C=C). ^{1}H-NMR(400MHz, DMSO-d_6): δ(*t*, 1.26-1.29ppm, 4H, -CH_2-CH_2-), δ(*d*, 3.70-3.72ppm, 4H, -CH_2-CH_2-), δ(*q*, 4.21-4.26ppm, 1H, -CH-), δ(*dd*, 6.83-6.85ppm, J=8, 2H+ 1H, Para Para System+C=CH) δ(*dd*, 7.79-7.81ppm, J=8, 2H, Para Para System) ^{13}C-NMR (400MHz, DMSO-d_6): δ(18.60ppm, -CH_2-CH_2-), δ(30.78ppm, -CH_2-CH_2-), δ(56.10ppm, -CH-), δ(125.43-134.35ppm, aromatic proton), δ(166.44ppm, C=O) δ(163.48ppm, C=N) δ(135.92ppm, C=CH).

2.2.1.1.3: 4-((2-Hydroxynaphthalen-1-yl)methylene)-2-(4-(trifluoromethyl)phenyl)

oxazol-5(4H)-one(3B). It reactions were used to create. A few drops of acetic anhydride, glycine(0.01mol, 0.75g), 2-hydroxy-1-naphthaldehyde(0.01mol, 1.72g), 4-((trifluoromethyl)benzoylchloride(0.01mol, 2.08g, 1.48ml), and sodium acetate (0.01mol, 0.8g). **m.p**.= 147-149^{0}C. **R_f** = 0.7. **Yield**= 96%, **IR** (*ῡ*, cm^{-1}, KBr disk). 3378(OH stretching vibration), 3065,3108(sp^2-CH stretching frequency), 1702(C=O), 1678(C=N), 1572(C=C). **^{1}H-NMR(400MHz, DMSO-d_6):** δ(*dd*, 8.08-8.10ppm, J=8, 2H, Para Para System), δ(*dd*, 8.41-8.43ppm, J=8, 2H, Para Para System), δ(*m*,7.63-8.20]ppm, 6H, aromatic proton), δ(*s*,9.39ppm,1H, C=CH), δ(*s*,10.28ppm,1H, OH). **^{13}C-NMR (400MHz, DMSO-d_6):** δ(122.55-143.72ppm, aromatic proton), δ(166.35 ppm, C=O), δ(165.53ppm, C=N), δ(137.91ppm, C=C). The mass spectra of (**3B**) showed the molecular ion peak [M+H]$^+$, at m/z=383.33 and the important fragmentation peaks at 57.1m/z, 95.1m/z, 121.1m/z, 149.0m/z, 173.1m/z, 201.2m/z, 227.1m/z, 253.2m/z, 279.2m/z, 311.3m/z, 339.3m/z, 359.3m/z & 382.3m/z,

2.2.1.1.4: 4-((2-Hydroxynaphthalen-1-yl)methylene)-2-(4-((trifluoromethyl)thio)p h enyl)oxazol-5(4H)-one(4B)**.**

It reactions were used to create. A few drops of acetic anhydride, glycine(0.01mol, 0.75g), 2-hydroxy-1-naphthaldehyde(0.01mol, 1.72g), 4-((trifluoromethyl)thio)benzoyl chloride(0.01mol, 2.40g, 1.66ml), and sodium acetate (0.01mol, 0.8g). **m.p**.= 199-200^{0}C. **R_f** = 0.9. **Yield**= 81%, **IR (ῡ, cm^{-1}, KBr disk).** 3379(OH stretching vibration), 3108,3062(sp^2-CH stretching frequency), 1702(C=O), 1678(C=N), 1572(C=C). **^{1}H-NMR(400MHz, DMSO-d_6):** δ(*dd*, 8.09-8.11ppm, *J*=8, 2H, Para Para System), δ(*dd*, 8.42-8.44ppm, *J*=8, 2H, Para Para System), δ(*m*,7.64-8.21ppm, 6H, aromatic proton), δ(s,9.40ppm,1H, C=CH), δ(*s*, 10.29ppm,1H, OH). **^{13}C-NMR(400MHz, DMSO-d_6):** δ(122.56-143.72ppm, aromatic proton), δ(166.35ppm, C=O), δ(165.53ppm, C=N), δ(137.91ppm, C=C). The mass spectra of (**4B**) showed the molecular ion peak [M+H]$^+$, at m/z=415.2 and the important fragmentation peaks at 51.2m/z, 75.1m/z, 95.1 m/z, 125.2m/z, 145.1m/z, 173.1m/z 355.2m/z, 212.2m/z, 233.2m/z, 256.2m/z, 281.2m/z, 320.2 m/z, 355.2m/z, 383.3 m/z & 405.2m/z.

2.2.1.1.4: 2,2'-(1,3-Phenylene)bis(4-((2-hydroxynaphthalen-1-yl)methylene)oxazol -5(4H)-one)(5B).

It reactions were used to create. A few drops of acetic anhydride, glycine(0.02mol, 1.5g), 2-hydroxy-1-naphthaldehyde(0.02mol, 3.44g), isophthaloyl dichloride(0.01mol, 2.03g), and sodium acetate(0.02mol, 1.6g). **m.p**.=148-150^{0}C. **R_f** =0.8. **Yield**=75%. **IR**(ῡ, cm^{-1}, KBr disk), 3330(OH stretching vibration), 3108,3067(sp^2-CH stretching frequency), 1765(C=O), 1655(C=N), 1591(C=C) **^{1}H-NMR(400MHz, DMSO-d_6).** δ(*m*,7.09-8.96ppm, 16H, aromatic proton), δ(*s*,8.61ppm,2H, C=CH), δ(*s*,9.33ppm,2H, OH). **^{13}C-NMR (400MHz, DMSO-d_6):** δ(115.69-150.07ppm, aromatic proton), δ(165.90ppm, C=O) δ(163.83 ppm, C=N), δ(150.74ppm, C=C).

Table(1):Physical data of mono-oxazolone B(1-5)

No.	m.p ^{0}C	R_f	Yield %	Colour	Solvent of recrystalization
1B	258-260	0.7	85	Dark-yellow	Benzene
2B	93-95	0.9	82	Red	Chloroform
3B	147-149	0.7	96	Red	Benzene
4B	199-200	0.9	81	Red	Chloroform
5B	148-150	0.8	75	Brown	Chloroform

2.2.2: General Procedures for the preparations of Oxazolone14.

2.2.2.1: Preparation method of Mono-oxazolone.

In general, mono-oxazolone is made by a porcelain mortar and pestle, glycine(0.01mol), derivatives aromatic aldehyde(0.01mol), derivatives benzoyl chloride(0.01mol), and fused sodium acetate (0.01mol) were combined for a few minutes in the presence of a few drops of acetic anhydride. The reaction mixture converted to solid after completion, as indicated by TLC, which was washed with cold water and recrystallized from ethanol to get the required azlactone. The oxazolone structures were validated by comparing their **m.p**., mixed. **m.p**., **TLC, IR, ^{1}H-NMR, ^{13}C-NMR & Mass** data to those in the literature. **Table(2):** Shows the physical properties of **mono-oxazolone.**

2.2.2.1.1: 4-(2,4-Dichlorobenzylidene)-2-(3-hydroxyphenyl)oxazol-5(4H)-one(6B).

It reactions were used to create. A few drops of acetic anhydride, glycine(0.01mol, 0.75g), 2,4-dichlorobenzaldehyde(0.01mol, 1.75g), 3-hydroxybenzoyl chloride(0.01 mol, 1.56g, 1.11ml) and fused sodium acetate (0.01mol, 0.8g). **m.p**.= 155-156^{0}C, **R_f** = 0.8. **Yield**=

64%. **IR**(ῡ, cm^{-1}, KBr disk). 3369(OH stretching vibration) 3061,3077, (sp^2-CH stretching frequency), 1705(C=O), 1621(C=N), 1583(C=C). **^{1}H-NMR (400 MHz, DMSO-d_6).** δ(*m*, 6.31-7.88ppm, 7H, aromatic proton) δ(*s*,7.20ppm, 1H, C=CH), δ(*s*,9.99ppm, 1H, OH). **^{13}C-NMR(400MHz, DMSO-d_6).** δ(109.77-149.09ppm, aromatic proton), δ(169.73ppm, C=O), δ(163.63ppm, C=N), δ(151.57ppm, C=C).

2.2.2.1.2: (E,Z)-4-(4-Hydroxy-3-methoxybenzylidene)-2-(4-nitrophenyl)oxazol-5 (4H)-one(7B)**.**

It reactions were used to create. A few drops of acetic anhydride, glycine(0.01mol, 0.75g), 4-hydroxy-3-methoxybenzaldehyde(0.01mol, 1.52g), 4-nitrobenzoyl chloride(0.01mol, 1.85g) and fused sodium acetate(0.01mol, 0.8g). **m.p**.= 119-120^{0}C. **R_f** = 0.6. **Yield**= 76%. **IR** (ῡ, cm^{-1}, KBr disk). 3393(OH stretching vibration), 3080,3115(sp^2-CH stretching frequency), 1699(C=O), 1637(C=N), 1604 (C=C). **^{1}H-NMR(400MHz, DMSO-d_6).** δ(*s*,3.83ppm,3H, -OCH_3), δ(*dd*, 8.15-8.17 ppm, *J*=8, 2H, Para Para System), δ(*dd*, 8.30-8.32ppm, *J*=8, 2H, Para Para), δ(*dd,dd, dd,* 6.94-7.43ppm, *J*=4,4,8, 3H, aromatic proton). For the ***Z*-isomer (35%):** 7.37(s, 1H, CH =C), for the ***E*-isomer (65%):** 7.38(s, 1H, CH=C), δ(*s*, 9.76ppm, 1H, OH). **^{13}C-NMR(400MHz, DMSO-d_6).** δ(55.63ppm,-OCH_3) δ(110.70-148.23ppm, aromatic proton), δ(165.98ppm, C=O), δ(153.11ppm, C=N), δ(150.04 ppm, C=C). The mass spectra of (**7B**) showed the molecular ion peak $[M+H]^+$, at m/z=340.29 and the important fragmentation peaks at 51.1m/z, 77.1m/z, 105.1m/z, 124.0m/z, 151.1m/z, 178.1m/z, 211.1m/z, 244.1m/z, 271.1m/z, 300.2m/z & 321.1m/z.

Table (2) physical data of mono-oxazolone B(6-7)

No.	m.p 0C	Rf	Yield %	Colour	Solvent of recrystalization
6B	155-156	0.8	64	yellow	Ethanol
7B	119-120	0.6	76	dark yellow	Ethanol

2.3. Hemolysis Assay of Mono-Bis oxazolone:

The hemolysis of oxazolone derivatives at different concentrations (5, 0.5, 0.05) mg/ml was determined using a red blood cell hemolysis assay released from RBCs. Blood samples from healthy individuals were acquired from the Al-Hussein Teaching Hospital's labs in AL-Muthanna. Each dilution was given a total volume of 0.8 ml in a test tube. Each tube was filled with human erythrocytes to a final volume of 1ml and incubated at 37^{0}C for 30 minutes. The mixtures were then centrifuged at 1000g for 5 minutes. The absorbance of the supernatant at 570 nm was measured using a UV-Vis spectrophotometer. Distilled water and DMSO were used as positive and negative controls, respectively. The hemolysis was calculated using the equation below. The experiment was carried out twice more.

HR (%) =(A sample - A negative control / A positive control - A negative control) X 100

Hemolysis (%) = (D_s − D_n) / (D_P − D_n) × 100

where D_s, D_n, and D_P are the absorbances of the sample, saline, and distilled water, respectively.

2.4: Antioxidant Activity of Mono-Bis oxazolone.

The synthesized mono-bis oxazolone derivatives concentrations and ascorbic acid (as a standard) were examined for the scavenging impact on 1,1-Diphenyl-2-Picry hydroxyl (DPPH) radical techniques, using a freshly created (0.004 w/v) DPPH methanol solution.was mixed with 1 mL oxazoloneDifferent concentrations of test sample solution (12.4, 50, 200, 400, 750, 800, and 1000 g/ml) were created. In the dark, the reaction solution was kept at 25°C for 30 minutes. A spectrophotometer UV was used to measure the absorbance at 517 nm. Ascorbic acid, which has antioxidant action, was used as a positive regulator. The percentage of free radicals inhibited was reported as a percentage of the total number of free radicals inhibited. The percentage of inhibition was calculated using following equation:

% DPPH scavenging activity = ($A_{control}$ − $A_{test\ sample}$ / $A_{control}$) × 100

2.5: Anti-bacteriological Activity of oxozolone compounds:

The antimicrobial activity of oxazolones compounds was determined using the micro broth dilution experiment with slight modifications. Consider the following:

2.5.1: Inoculum Preparation:

A sterile Mueller-Hinton Broth was inoculated with a loop of bacterial colonies from an overnight solid medium culture (MHB). Prior to antimicrobial testing, broth inoculums were cultured at 37^{0}C for 24 hours. The final inoculum size was (1×10^6 CFU/ml) after the broth inoculum was adjusted to the 0.5 McFarland tube and diluted to a ratio of 1:100.

2.5.2: Broth Micro dilution:

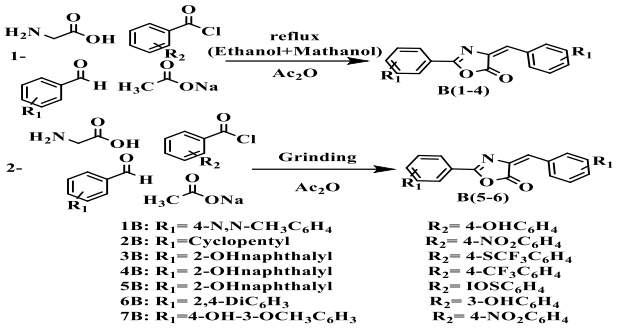
The chemical compounds were first dissolved in 10% DMSO, then serially diluted two times in a liquid growth medium (MHB) with 300-0.585 mg/ml of each compound. Except for the 12th well, 100 l of each compound dilution was applied to each well of 96 well micro titer plates after shaking (growth control, without the compound).Then, each well is inoculated with 100 µl of a microbial inoculum (1×10^6 CFU/ml), except of the 11th well (sterility control, without bacteria), and incubated at 37±2^{0}C. MIC was defined as the lowest concentration of compound that inhibits bacteria after 24h. Absorbance reading (by ELISA reader) at 630 nm wavelength for each plate was measured pre- and post-incubation at 37^{0}C for 24 hours. According to calculation of the Bacterial growth inhibition was achieved by the following equation:

$$\text{Percentage growth inhibition} = \frac{\text{OD of control}}{\text{OD of control - OD of test}} \times 100$$

3. Results and Discussion

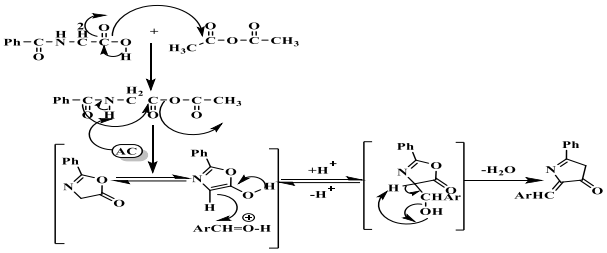
Chemistry. Novel oxazolone compounds can be synthesized in just one step. New oxazolone compounds were produced in a number of techniques. The first method [11] way is to use Glycine, aromatic aldehyde, benzoyl chloride, sodium acetate, and a few drops of acetic anhydride were used to make oxazolone compounds, which were refluxed for 9-12 hours in the presence of the solvent [Methanol+ Ethanol]. The second method [12] Synthesis of oxazolone compounds in Glycine, aromatic aldehyde, benzoyl chloride, and fused sodium acetate were mixed for a few minutes in the presence of a few drops of acetic anhydride in a porcelain mortar and peinstle. for free-Solvent as shown in

Scheme1.



Scheme1

The proposed mechanism [13] for their formations Mono-Bis oxazolone was shown as below in **Schemes2.**



Schemes2

IR, ^{1}H-NMR, ^{13}C-NMR, & Mass spectrum data were used to determine the structures of the Mono-Bis oxazolone.

3.1: Hemolysis Study

Hemolysis is an important metric for estimating anticancer agents that have been produced. The amount of hemoglobin discharged can be measured quantitatively to determine the risk of RBC destruction [14]. When the hemolysis ratio is less than 10%, the produced anticancer drugs can be administered intravenously. The hemolysis ratio percentages of all Mono-Bis oxazolone were smaller at a greater concentration (0.5 mg/ml) according to the results **(Table3).** The percentages of hemolysis ratios in all concentrations were less than 4%. This finding reveals that manufactured Mono-Bis oxazolone compounds for intravenous injection have high blood safety [15].

Table3. The results of a hemolysis experiment for oxazolone compounds B(1-7)

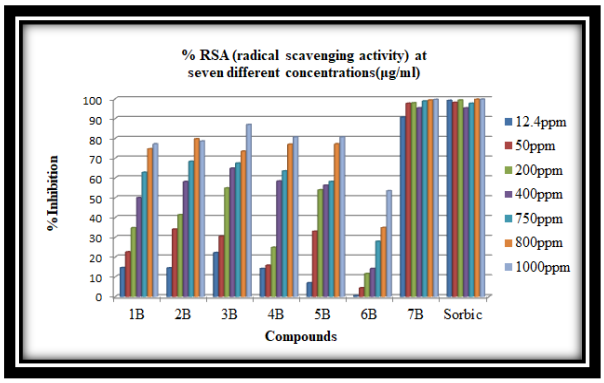
Compound code	HR (%)		
	0.005 mg/ml	0.05 mg/ml	0.5 mg/ml
1B	0.07	0.19	1.94
2B	0.08	0.18	1.81
3B	0.04	1.47	2.69
4B	0.73	1.53	2.45
5B	0.79	1.13	2.84
6B	0.28	1.73	2.99
7B	0.07	0.58	2.72

3.2: Antioxidant Activity

When the pace of free radical synthesis surpasses the rate of oxidative stress, a damaging process occurs, which can damage cell components such as lipids, proteins, and DNA. As a result, defensive mechanisms that deconstruct and stabilize free radicals safeguard all species from free radical attacks. Antioxidants are chemicals that neutralize free radicals, preventing oxidative cell damage. The potential of manufactured compounds to behave as free radical scavengers and to estimate antioxidant activity is frequently measured using DPPH, a standard and stable free radical. The amount of DPPH is lowered. through the acceptance of an electron or a molecule of hydrogen [16]. The Mono-Bis oxazolone's capacity to scavenge radicals in vitro DPPH radical scavenging assays are used to examine it. The reduced activity of the samples was determined by changing the color of the DPPH solution from deep purple to yellow. As indicated in **(Table4),** the antioxidant activity of the samples is calculated and compared to the standard antioxidant ascorbic acid. The DPPH radical was used to calculate the free radical scavenging special effects of all the produced compounds at various concentrations (1000, 800, 750, 400, 200, 50, and 12.4) ppm. On the other hand, it is widely established that organic compounds containing electron donating groups (amine, methoxy, and hydroxyl) can act as free radical scavenging agents. The percentages of samples that are inhibited range from 53.49% to 99.60%. In a 1000 ppm concentration of vitamin C, 99.77% inhibition was observed. In comparison to all of the test chemicals in 1000 ppm, compound (7B) showed the highest level of inhibition at 99.60%. as compared with other compounds due to electron donating group OH,OCH_3. Furthermore, all samples with concentrations less than 1000 ppm showed decreased inhibition. In 1000 ppm, compound(6B) showed the least inhibition (53.49%). When compared to vitamin C, all of the substances examined had varying antioxidant properties.

Table 4. The inhibitory values displayed by the test Mono-Bis oxazolone compounds B(1-6)

% RSA (radical scavenging activity) at seven different concentrations(µg/ml)							No
12.4ppm	50ppm	200ppm	400ppm	750ppm	800ppm	1000ppm	
14.65	22.63	34.81	50.00	62.79	74.65	77.21	1B
14.47	34.09	41.40	58.14	68.37	79.77	78.60	2B
22.23	30.47	54.88	64.88	67.44	73.49	86.98	3B
14.19	15.81	24.88	58.37	63.49	76.98	80.70	4B
6.98	33.02	53.95	56.28	58.14	77.21	80.70	5B
0.47	4.42	11.63	14.19	27.91	34.88	53.49	6B
90.70	97.67	97.91	95.35	98.84	99.30	99.60	7B
99.12	98.14	99.33	95.35	97.67	99.72	99.77	Sorbic



Fig(1):Radical scavenging activity (RSA) for the synthesized compounds

3.3: Antibacterial Activity.

The antibacterial activity of the synthesized compounds

(3B, 4B & 7B) [17] were tested against (*Staphylococcus aureus*) The anti-bacterial activity of some of the prepared compounds has been evaluated by microbroth dilution method were prepared in differented concentrations as (400 mg\ml, 200 mg\ml, 100, 50 mg\ml, 25 mg\ml, 12.5 mg\ml, 6.25 mg\ml, 3.125 mg\ml, 1.56mg\ml, 0.781 mg\ml). And it is shown by the data of the obtained tests mentioned in Tables(5) & Fig(2,3,4). In general, the compounds give different inhibiting, according to the type of rings, the nature of compensation, the concentration of the material, and the type of bacteria under test [18]. From the data obtained, in the oxazolone derivatives, we note that the derivative (7B) have the highest inhibitory power as compared to other derivatives in the same kind against Staphylococcus aureus (gram +ve). While we note that the derivative (4B) have the lowest inhibitory power as compared to other derivatives in the same kind against Staphylococcus aureus (gram +ve).

(Table5): Growth inhibition percentages of Staphylococcus aureus against a different concentrations of the eight studied compounds. Data represent the mean ± SD

Concentration	Comp. H=(3B)		Comp. F=(4B)		Comp. G=(7B)	
	Mean	S.D.	Mean	S.D.	Mean	S.D.
400 mg\ml	100	0	100	0	100	0
200 mg\ml	100	0	100	0	100	0
100 mg\ml	100	0	99.6042	0.43994	100	0
50 mg\ml	100	0	95.2866	1.01118	100	0
25 mg\ml	75.9865	3.0089	93.1043	1.00107	85.5803	1.497
12.5 mg\ml	26.3943	2.36146	54.5329	1.25035	40.1349	1.90233
6.25 mg\ml	8.3797	1.09647	27.4832	2.13372	29.8824	1.58049
3.125 mg\ml	6.5854	1.36107	16.4679	1.04241	20.6902	1.4414
1.56 mg\ml	3.2043	1.0417	10.5619	1.39847	16.1076	1.1729
0.781 mg\ml	0	0	3.6217	0.73107	9.3913	1.34266

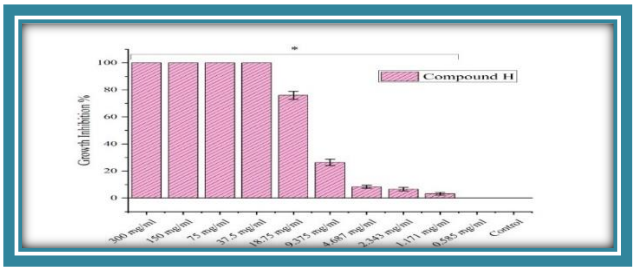


*Fig. (2): Growth inhibition of Staphylococcus aureus caused by different concentration of the compound H=(3B). * represent a significant difference at p≤0.05 between the four purified compound concentrations and control. Data represent the mean ± SD.*

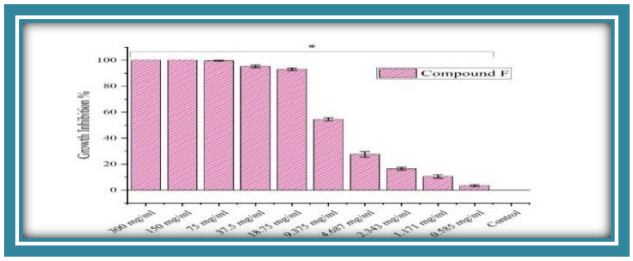


*Fig. (3): Growth inhibition of Staphylococcus aureus caused by different concentration of the compound F=(4B). * represent a significant difference at p≤0.05 between the four purified compound concentrations and control. Data represent the mean ± SD.*

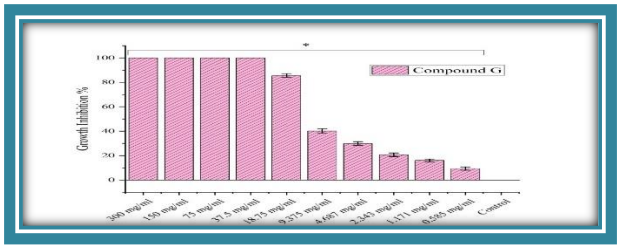


*Fig. (4): Growth inhibition of Staphylococcus aureus caused by different concentration of the compound G=(7B). * represent a significant difference at p≤0.05 between the four purified compound concentrations and control. Data represent the mean ± SD.*

4. Conclusions

The novel oxazolone compounds were produced in high yields (64-85%) and showed antioxidant and cytotoxicity properties. FT-IR ^{1}HNMR and ^{13}CNMR were used to prove the chemical structure of the produced molecules. The safety of oxazolone use inside the body was demonstrated in a hemolysis study. Oxazolone has a significant DPPH radical antioxidant activity, according to antioxidant experiments on synthetic compounds.

5. Recommendations

Based on the findings, we recommend the following substances for usage as medicines due to their excellent efficacy.

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