*Iranian Journal of Organic Chemistry Vol. 16, No. 3 (2024) 3847-3854*

# $Fe_3O_4$MNPs promoted green synthesis of 1,3-oxazole derivatives: Study of antimicrobial and antioxidant activity

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Received:May 2024; Revised: June 2024; Accepted: July 2024

**Abstract:** In this research bio-Fe3O4-MNPs (iron-oxide magnetic nanoparticles) promoted synthesis of 2-thioxo-1,3-oxazole derivatives using multicomponent reaction of α-bromo ketones, alkyl (aryl) isothiocyanates, sodium hydride and catalytic amount of Fe3O4 MNPs in water at room temperature in good yields. Also, Fe3O4-MNPs were generated using Orange peel water extract as green procedure that reduce the ferric chloride solution. The nanoparticles that is generated via biosynthesis method have potentially valuable in different purposes such as organic synthesis. In addition, for study of antioxidant ability of some synthesized thioxo-1,3-oxazoles, diphenyl-picrylhydrazine (DPPH) radical trapping and power of ferric reduction testes are employed. Among studied thioxo-1,3-oxazoles, 4b have good power for radical trapping and reduction activity than to standard antioxidant such as BHT and TBHQ. In addition, the antimicrobial activity of some thioxo-1,3-oxazoles was studied employing the disk diffusion test on Gram-positive bacteria and Gram-negative bacteria. The results of disk diffusion test showed that compound 4a, 4b, 4d and 4f prevented the bacterial growth.

**Keywords:**2-thioxo-1,3-oxazole, Orange peel extract, Fe3O4-MNPs, Alkyl (aryl) isothiocyanates, Alkyl bromides, antioxidant ability.

## Introduction

The multi-component reactions used for synthesis of heterocyclic compounds in the last decade. Organic compounds in multi-component reactions (MCRs), were generated in a few steps or in a one-pot procedure [1-3].Also, the green chemistry represent chemical procedures that decrease or avoid the application and production of hazardous chemicals for the environment. Organic solvents that are needed for performing some organic reactions are often toxic and expensive. For this reason, elimination of these solvents is a suitable work for nature. Therefore, performing reactions in water as a solvent has received manyattentions in recent years [4]. Water is an inexpensive solvent which is available in large amounts and can increase the rate of organic reaction even for compounds that are insoluble in water. Separation of the product in water was also done by simple purification [5-7].Among the heterocycles, the furo-oxazole motif has used as a synthetic precursor for the chemical syntheses of some rare sugars and antisense oligonucleotides [8, 9]. It is reported that important groups of compounds with the furo-oxazole skeleton are prodrugs of caspase inhibitors with apoptosis-regulating activity [10].However, there are very limited methods available to synthesize of these compoundssome methodologies are existed for the synthesis of this type of heterocycles [11]. Therefore, the search for new synthetic strategies leading to these compounds from simple starting materials in a sustainable and atom-economical fashion is of continued interest of our group. Also, the finding of novel synthetic methods towards oxazole derivatives is a part of continued interest for organic chemists. The preparation of oxazole derivatives are

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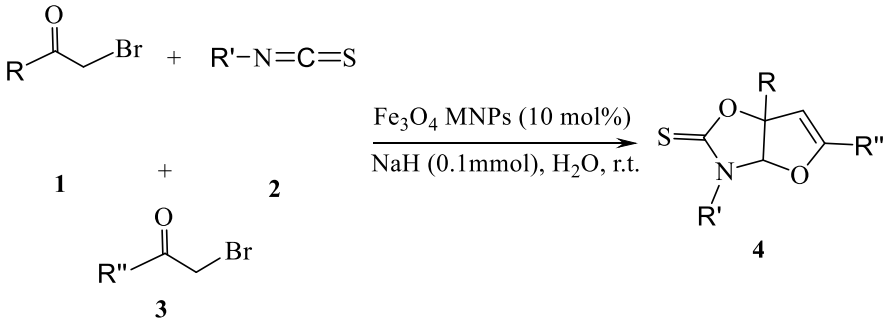
attractive due to having many biological activity such as antibacterial, anti-fungal, anti-tubercular and anti-inflammatory activitiesin addition to their applications as important precursors in many convenient synthetic transformations [12]. Alsooxazoles are used as shining compounds and textiles fluorescent whitening factors and very important in dye chemistry [13, 14]. Fused bicyclic oxygen-containing heterocycles are embodied in a wide range of natural products, modified sugar derivatives, and important bioactive molecules.

Another topic in this work is study of antioxidant activity of some synthesized compounds. Usually, the compounds that because of their reductive properties and chemical structure have antioxidant activity employ as transitional metals chelators and removing the negative effect of free radicals. These compounds with antioxidant activity could be prevent or reduce many diseases such as cardiovascular, inflammatory bowel syndrome, cancer, ageing, and Alzheimer.

At present, bacteria that is stable in the presence of drug have created substantial problems in the performance of many communicable diseases. Therefore, discovering new ways to fight against these pathogens are important. For this reasone, recent studies have focused on the study of the antibacterial effects of new synthesized compounds. Herein, in continuation of our studies for discovering new procedure for synthesis of important heterocyclic compounds with biological activity [15, 16] in this work functionalized oxazoles**4** was produced in excellent yields from the reaction of alkyl bromides **1**, isothiocyanate **2**, alkyl bromides **3**, sodium hydride and catalytic amount of $Fe_3O_4$ MNPs (10 mol%) in water at room temperature (Scheme **1**).




| 4 | R | R | R" | Yied % of **4** |
|---|---|---|---|---|
| **a** | $CO_2Et$ | 4-MeO-$C_6H_4$ | $CO_2Et$ | 95 |
| **b** | $CO_2Et$ | 4-$NO_2$-$C_6H_4$ | $CO_2Et$ | 90 |
| **c** | $CO_2Et$ | $^t$Bu | $CO_2Et$ | 92 |
| **d** | 4-MeO-$C_6H_4$ | $^t$Bu | 4-MeO-$C_6H_4$ | 92 |
| **e** | 4-Me-$C_6H_4$ | 4-MeO-$C_6H_4$ | 4-Me-$C_6H_4$ | 87 |
| **f** | 4-Br-$C_6H_4$ | 4-$NO_2$-$C_6H_4$ | 4-Br-$C_6H_4$ | 83 |

**Scheme 1.** Synthesis of oxazole derivatives **4**

## Results and Discussion

### Chemistry

For achieving the best condition for reaction, we select synthesis of compound **4a** as sample reaction. Synthesis of oxazole 4a was performed in excellent yields from the reaction of ethyl bromopyruvate **1a**, 4-methoxy phenyl isothiocyanate **2a**, ethyl bromopyruvate **3a** and sodium hydride. For this purpose, catalyst, solvent and temperature changed and yield of 4a as sample reaction was achieved. For investigation of catalyst, ZnO-NPs, $TiO_2$-NPs, KF/CP-NPs and $Fe_3O_4$-MNPs are applied as catalyst. Among them $Fe_3O_4$-MNPs is the best due to green preparation, easy separation of catalyst from the mixture of reaction and easy purification of product. Several solvents such as $CH_2Cl_2$, $CH_3CN$, toluene and $H_2O$ are applied for these reactions. Among them $H_2O$ is the best because of simple removal of product via filtration. The outcomes of optimization are displayed in Table 1.

For the synthesis of $Fe_3O_4$-MNPs, the aqueous extract of orange peel was used. For confirming the structure of $Fe_3O_4$-MNPs, field emission scanning electron

microscopy (FESEM) (Figure **1**) and X-Ray nanostructure. FESEM image of $Fe_3O_4$MNPs display spherical morphology with uniform-sized particles.

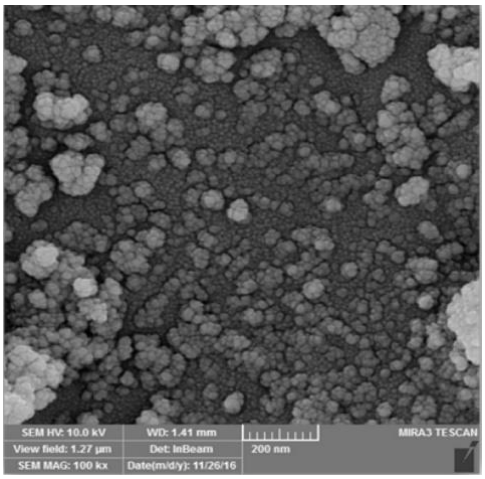
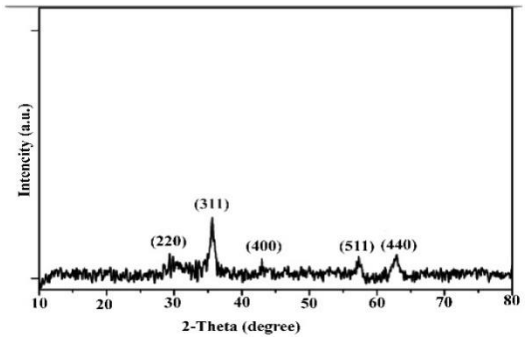



**Figure 1.** FESEM images of $Fe_3O_4$-MNPs

**Table 1:** Achieving the best conditions for preparation of **4a**

| Entry | Catalyst | Solvent | Temp. (ºC) | Time (h) | Yield (%)[a] |
|---|---|---|---|---|---|
| 1 | ----- | $CH_2Cl_2$ | r.t. | 6 | 10 |
| 2 | ----- | $H_2O$ | r.t. | 12 | 50 |
| 3 | ----- | $H_2O$ | reflux | 8 | 50 |
| 4 | | $CH_3CN$ | r.t. | 8 | 37 |
| 5 | ----- | $CH_3CN$ | reflux | 8 | 37 |
| 6 | ----- | toluene | r.t. | 7 | 45 |
| 7 | ----- | toluene | reflux | 7 | 45 |
| 8 | KF/CP NPs | $CH_2Cl_2$ | r.t. | 8 | 35 |
| 9 | KF/CP NPs | $H_2O$ | r.t. | 10 | 65 |
| 10 | KF/CP NPs | $H_2O$ | reflux | 7 | 65 |
| 11 | KF/CP NPs | $CH_3CN$ | r.t. | 7 | 45 |
| 12 | KF/CP NPs | $CH_3CN$ | reflux | 7 | 45 |
| 13 | KF/CP NPs | toluene | r.t. | 7 | 50 |
| 14 | ZnO (NPs) | $H_2O$ | r.t. | 10 | 47 |
| 15 | ZnO (NPs) | $H_2O$ | reflux | 8 | 48 |
| 16 | ZnO (NPs) | $CH_3CN$ | r.t. | 8 | 70 |
| 17 | ZnO-NR | $CH_2Cl_2$ | r.t. | 12 | 37 |
| 18 | ZnO-NR | $H_2O$ | r.t. | 10 | 45 |
| 19 | ZnO-NR | toluene | r.t. | 8 | 60 |
| 20 | ZnO-NR | $CH_3CN$ | r.t. | 10 | 80 |
| 21 | $TiO_2$-NPs | $CH_2Cl_2$ | r.t. | 12 | 45 |
| 22 | $TiO_2$-NPs | $H_2O$ | r.t. | 8 | 50 |
| 23 | $TiO_2$-NPs | $CH_3CN$ | r.t. | 10 | 48 |
| 24 | $TiO_2$-NPs | toluene | r.t. | 8 | 65 |
| 25 | **$Fe_3O_4$-MNPs** | **$H_2O$** | **r.t.** | **6** | **87** |
| 26 | $Fe_3O_4$-MNPs | $CH_3CN$ | r.t. | 8 | 76 |
| 27 | $Fe_3O_4$-MNPs | toluene | r.t. | 8 | 67 |

The X-ray diffraction model of the $Fe_3O_4$ nanoparticles is showed in Figure 2. The peaks at 2θ = 30.4°, 35.6°, 43.1°, 57.5° and 62.7° can be showed to (220), (311), (400), (511) and (440) It was confirmed to cubic inverse spinel structure compared well with Diffraction (XRD) (Figure **2**) image are taken for




the JCPDS card no. 19–0629. The half-value size of the $Fe_3O_4$ nanoparticles is calculated 16 nm.

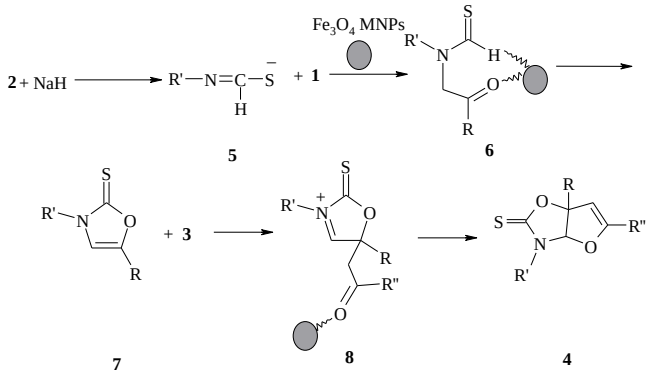
**Figure 2.** XRD spectra of $Fe_3O_4$ nanoparticles

Without employing catalyst, these reactions have low yield and busy mixture. The synthesis of compound 4a as sample reaction have alike yield in the presence of ZnO-NPs and $Fe_3O_4$ MNPs (entry 20 and entry 30) but removal of catalyst from the mixture of reaction after completing of reaction is comfortable in present of $Fe_3O_4$ MNPs. Structures of **4a–4f** are confirmed by IR, $^1$H NMR, $^{13}$C NMR mass spectra. For example, the $^1$H NMR spectrum of **4a** exhibits one singlet at δ = 3.78 ppm for methoxy protons and two singlets at δ = 6.85, 7.62 ppm for methine proton in association with signals for aromatic section. The resonance of carbonyl and thionyl group was appeared at 163.2 (C=O), 164.2 (C=O), 187.4 (C=S) ppm respectively in 13C NMR spectra of 4a. A suggested mechanism for this reaction is showed in Scheme 2. The reaction begins with creation of intermediate **5** from the reaction of isothiocyanate **2** and sodium hydride that is activated by $Fe_3O_4$ MNPs as catalyst. Alkyl bromides 1 react with intermediate **5** and produced intermediate **6** that intermolecular cyclization of intermediate **6** generate compounds **7**. Compounds **7** react with compounds 3 as nucleophile and produce intermediate **8.** Finally, intermolecular cyclization of intermediate 8 generates compounds **4**.

## Study of antioxidant activity employing Diphenyl-2-picrylhydrazyl (DPPH)

For determination of antioxidant activity of some synthezied compounds and their antioxidant property in foods and biological systems as well as power of compounds to take free radicals, DPPH radical trapping experiment is widely used. In this experiment, the DPPH radical takes the hydrogen atom (or one electron) of synthezied compounds **4a-4d** and gives an

evaluation of antioxidant activity basis of free radical trapping. The absorption of DPPH radical was observed area 517 nm but when DPPH radical is reduced by an antioxidant or a radical species its absorption decreases. As shown from the results, free radical trapping activity of compounds **4a-4d** is excellent and lower than BHT and TBHQ (Figure 3). Normally, the DPPH scavenging ability of these compounds was attained TBHQ>BHT>**4b>4a>4d>4c** respectively. The free radical trapping power had been enhanced from 200 to 1000 ppm. So, by rising concentration in all samples, the free radical activity was raised. For instance, compound **4b** with a concentration of 1000 ppm had 88.53% inhibition while a concentration of 200 ppm of compound **4b** was exhibited 29.42% free radical inhibition.




**Scheme 2.** Proposed mechanism for Synthesis of 1, 3-oxazole derivatives

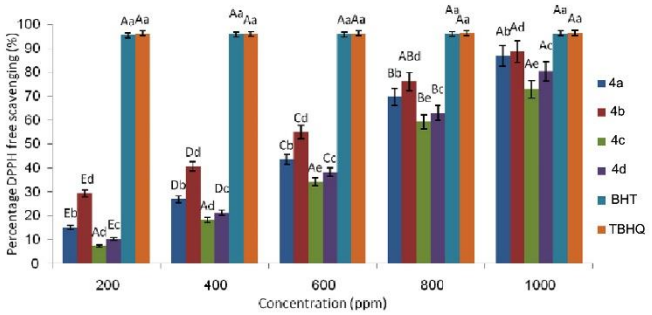



**Figure 3.** Radical scavenging activity (RSA) of compounds **4a-4d**.

## Ferric ions ($Fe^{3+}$) reducing potential (FRAP)

Reducing power of the synthesized compounds was determined by calculating of the exchange amount of $Fe^{3+}$/ferricyanide complex to the $Fe^{2+}$/ferrous form at 700 nm. The reducing power of compounds **4a-4d** compared with synthetic antioxidants (BHT and TBHQ) are showed in Figure 4. The bigger reducing power means higher absorbance of the compounds. The reducing activity order of compounds **4a-4d** was as following: TBHQ>BHT>**4b>4a>4d>4c** (Figure 4). In all them, the increasing concentration was enhanced ferric ions reducing power. Compounds **4b** show very good reducing activity compared to standards (BHT and TBHQ).

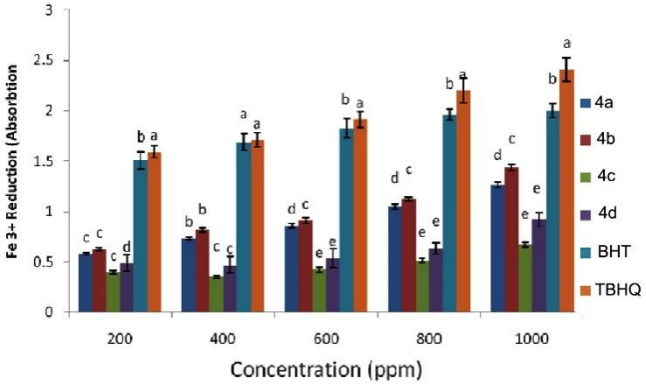



**Figure 4.** Ferric ions ($Fe^{3+}$) reducing antioxidant power (FRAP) of compounds **4a-4d**.Values in the same concentration followed by different letters are significantly different ($P$<0.05)

## Analysis of the antibacterial activity of synthesized compounds

Also, a comparison between the activity of our synthesized compounds with Streptomycin and Gentamicin as standard drug was discussed. The results of the antimicrobial activity of some synthezized compounds on bacterial species are shown in Table 2. The present study indicated that the type of bacteria and concentration of compounds are effective on the diameter of the inhibition zone. It is apparent from the data listed in Table 2, the antimicrobial activity of the most synthesized compounds **4a, 4b, 4d** and **4f** were good active against Gram positive bacteriaandGram negative bacteria So that the diameter of the inhibition zone of compounds has the maximum effect on *Escherichia coli*.

**Table 2.** The antibacterial activity of the tested compounds

| ***Compounds*** | ***Staphylococcus aureus* (+)** | ***Bacillus subtilis* (+) (PTCC 1023)** | ***Bacillus cereus* (+) (PTCC 1079)** | ***Pseudomonas aurignosa*(-)** | ***Escherichia Coli* (-)** | ***Klebsiella pneumoniae* (-) (PTCC 1053,** |
|---|---|---|---|---|---|---|

| | (PTCC 1431,112) | | | (PTCC 1430) | (PTCC 1399, 1533) | 1291) |
|---|---|---|---|---|---|---|
| **4a** | **17** | **20** | **19** | **17** | **23** [a] | **20** |
| **4b** | **16** | **18** | **17** | **15** | **20** | **18** |
| **4c** | --- | 10 | --- | ---- | 8 | 12 |
| **4d** | **17** | **19** | **15** | **16** | **20** | **21** |
| **4e** | 8 | 10 | 8 | 9 | 10 | 12 |
| **4f** | **18** | **20** | **18** | **17** | **21** | **19** |
| **4g** | 9 | 5 | 11 | -- | 7 | ---- |
| **Streptomycin** | 19 | 23 | 22 | 20 | 24 | 22 |
| **Gentamicin** | 22 | 20 | 20 | 19 | 21 | 21 |

[a]zone of inhibition in diameter in mm

## Conclusion

In conclusion, the reaction of α-bromo ketones, isothiocyanate and sodium hydride in the presence of catalytic amount of $Fe_3O_4$ MNPs in water generate thioxo-1,3-oxazole derivatives in good yields. Some advantages of performing these reactions with present procedure are carrying out these reactions in water as green solvent and simple removal of catalyst. For this reason these compounds are an interesting alternative to the complex multistep approaches. as well, green preparation, excellent yields, simple procedure, easy removal of catalyst from the mixture of reactions are the advantages of these reactions. Also, compound **4b** was shown a very good radical trapping activity and reducing activity relative to standards (BHT and TBHQ) by investigation of antioxidant activity. Moreover, the antimicrobial activity of some synthesized compounds was proved employing the disk diffusion test on Gram-positive and negative bacteria. The obtained results of disk diffusion test showed that compound **4a, 4b, 4d** and **4f** prevented the bacterial growth. Moreover, easy workup of catalyst and product, performing reactions in water and reusability of catalyst makes this method as an interesting option to other approaches.

## Experimental

### General

All of starting materials and solvents for these reactions and preparation of $Fe_3O_4$-MNPs and were used with any further purification. The $Fe_3O_4$ MNPs was prepared according to literature. An IR spectrum (KBr medium) for synthesized compounds was given by Shimadzu IR-460 spectrometer. Also, NMR spectra were achieved by a Bruker DRX-500 AVANCE spectrometer at 500 MHz for $^1$H-NMR and 125 MHz for $^{13}$C-NMR in $CDCl_3$ employing TMS as internal standard. Mass spectra (electron impact ionization) were taken by a Finnigan MAT 8430 spectrometer operating at an ionization potential 70 eV. The Heraeus CHN–O-Rapid analyzer was used for elemental analyses of products. Melting points are measured on an Electrothermal 9100 apparatus. The shape of $Fe_3O_4$ nanoparticles was confirmed by SEM image employing a Holland Philips XL30 microscope. Crystalline structure of Fe3O4 MNPs was confirmed by XRD analysis at room temperature using a Holland Philips Xpert X-ray powder diffractometer.

### Preparation of $Fe_3O_4$-MNPs

After dried Orange peel (10 gr) it was poured in 200 ml water at 80$^o$C. After 2 h, the mixture was filtered and water essential oil was applied for preparation of $Fe_3O_4$-MNPs as following. 100 ml Orange peel water extract was moved to a 250 and then $FeCl_3$ (2 mmol), $FeCl_2$ (1 mmol) were added two-neck round bottom flask. The solution of $NH_4OH$ (9 M, 10 ml) was added gently into the mixture of reaction under N2 atmosphere for 1 h at ambient temperature. The black color precipitate was produced and separated from mixture of reaction by external magnetic field and cleaned with water for several times as well as dried in oven at 80 $^o$C for 24 h.

### A general way to prepare of compounds 4

The isothiocyanate **2** (2 mmol) and α-bromo ketones **1** (2 mmol) was mixed together in water and stirred by

magnetic stirrer. After one minute sodium hydride (0.1 mmol) and $Fe_3O_4$ MNPs (10 mol %) was added to previouse mixture at room temperature. Then after 3h α-bromo ketones **3** (2 mmol) were added to previous mixture. After completion of the reaction (5 h; TLC control (hexane–AcOEt, 6:1), the removal of $Fe_3O_4$ MNPs were carried out by external magnet and the solid was extracted by filtration. The solide was disolved in ethyl acetate (2 mL) and purified by column chromatography (6:1 hexane/EtOAc) to afford pure title compound. After purification of compounds **4**, it was washed with diethyl ether several times to afforded pure title compounds.

*Diethyl 3-(4-methoxyphenyl)-2-thioxo-3,3a-dihydro-furo[2,3-d][1,3]-oxazole-5,6a(2H)-dicarboxylate* **(4a):**

Yellow powders; mp 115-117 °C, Yield: 0.75 g (95%). IR (KBr) (νmax/cm-1): 1739, 1692, 1587, 1467, 1374 and 1284 cm-1. 1H NMR: δ 1.23 (3 H, t, 3J = 7.3 Hz, Me), 1.32 (3 H, t, 3J = 7.3 Hz, Me), 3.78 (3 H, s, MeO), 4.23 (2 H, q, 3J = 7.3 Hz, CH2O), 4.34 (2 H, q, 3J = 7.3 Hz, CH2O), 6.85 (1 H, s, CH), 6.93 (2 H, d, 3J = 7.8 Hz, 2 CH), 7.24 (2 H, d, 3J = 7.8 Hz, 2 CH), 7.62 (1 H, s, CH) ppm. 13C NMR: δ 13.8 (Me), 14.2 (Me), 55.6 (MeO), 61.4 (CH2O), 62.5 (CH2O), 87.2 (C), 100.3 (CH), 115.8 (2 CH), 117.2 (CH), 129.6 (2 CH), 137.2 (C), 142.3 (C), 158.6 (C), 163.2 (C=O), 164.2 (C=O), 187.4 (C=S) ppm. MS, m/z (%): 393 (M+, 15), 348 (64), 45 (100).

*Diethyl 3-(4-nitrophenyl)-2-thioxo-3,3a-dihydro-furo[2,3-d][1,3]-oxazole-5,6a(2H)-dicarboxylate* **(4b):**

Yellow powders; mp 145-147 °C, Yield: 0.73 g (90%). IR (KBr) (νmax/cm-1): 1742, 1695, 1586, 1475, 1392 and 1294 cm-1. 1H NMR: δ 1.25 (3 H, t, 3J = 7.4 Hz, Me), 1.36 (3 H, t, 3J = 7.4 Hz, Me), 4.12 (2 H, q, 3J = 7.4 Hz, CH2O), 4.28 (2 H, q, 3J = 7.4 Hz, CH2O), 7.12 (1 H, s, CH), 7.41 (2 H, d, 3J = 7.6 Hz, 2 CH), 7.68 (1 H, s, CH), 8.06 (2 H, d, 3J = 7.6 Hz, 2 CH) ppm. 13C NMR: δ 13.5 (Me), 14.0 (Me), 61.6 (CH2O), 62.8 (CH2O), 87.3 (C), 101.4 (CH), 116.7 (2 CH), 118.3 (CH), 128.7 (2 CH), 140.3 (C), 142.5 (C), 147.4 (C), 163.5 (C=O), 164.6 (C=O), 189.3 (C=S) ppm. MS, m/z (%): 408 (M+, 10), 363 (78), 45 (100).

*Diethyl 3-(tert-butyl)-2-thioxo-3,3a-dihydro-furo[2,3-d][1,3]-oxazole-5,6a(2H)-dicarboxylate* **(4c):**

Pale yellow powders; mp 97-99°C, Yield: 0.63 g (92%). IR (KBr) (νmax/cm-1): 1738, 1695, 1587, 1486, 1397 a nd 1295 cm-1. 1H NMR: δ 1.28 (9 H, s, Me3C), 1.32 (3 H, t, 3J = 7.4 Hz, Me), 1.40 (3 H, t, 3J = 7.4 Hz, Me), 4.16 (2 H, q, 3J = 7.4 Hz, CH2O), 4.25 (2 H, q, 3J = 7.4 Hz, CH2O), 6.94 (1 H, s, CH), 7.73 (1 H, s, CH) ppm. 13C NMR: δ 13.8 (Me), 14.5 (Me), 29.3 (Me3C), 55.7 (Me3C), 61.8 (CH2O), 63.4 (CH2O), 85.7 (C), 96.3 (CH), 119.2 (CH), 143.4 (C), 162.8 (C=O), 165.3 (C=O), 190.2 (C=S) ppm. MS, m/z (%): 343 (M+, 15), 286 (84), 57 (100), 45 (100).

*3-(tert-butyl)-5,6a-bis(4-methoxyphenyl)-3a,6a-dihydro furo[2,3-d][1,3]-oxazole-2(3H)-thione* **(4d):**

Yellow powders; mp 138-140 °C, yield: 0.76 g (92%). IR (KBr) (νmax/cm-1): 1698, 1598, 1487, 1376 and 1295 cm-1. 1H NMR: δ 1.32 (9 H, s, Me3C), 3.78 (3 H, s, MeO), 3.83 (3 H, s, MeO), 6.93 (1 H, s, CH), 7.12 (2 H, d, 3J = 7.8 Hz, 2 CH), 7.26 (2 H, d, 3J = 7.6 Hz, 2 CH), 7.43 (2 H, d, 3J = 7.7 Hz, 2 CH), 7.68 (2 H, d, 3J = 7.7 Hz, 2 CH), 7.75 (1 H, s, CH) ppm. 13C NMR: δ 29.3 (Me3C), 55.3 (Me3C), 56.3 (MeO), 57.2 (MeO), 89.3 (C), 98.3 (CH), 107.5 (CH), 112.3 (2 CH), 113.5 (2 CH), 128.2 (2 CH), 129.4 (2 CH), 131.2 (C), 131.8 (C), 155.2 (C), 157.3 (C), 159.6 (C), 191.2 (C=S) ppm. MS, m/z (%): 411 (M+, 20), 354 (68), 57 (100).

*3-(4-methoxyphenyl)-5,6a-bis(4-methylphenyl)-3a,6a-dihydrofuro[2,3-d][1,3]-oxazole-2(3H)-thione* **(4e):**

Yellow powders; mp 163-165 °C, yield: 0.76 g (87%). IR (KBr) (νmax/cm-1): 1695, 1587, 1464, 1387 and 1293 cm-1. 1H NMR: δ 2.25 (3 H, s, Me), 2.37 (3 H, s, Me), 3.85 (3 H, s, MeO), 6.85 (1 H, s, CH), 6.96 (2 H, d, 3J = 7.6 Hz, 2 CH), 7.08 (2 H, d, 3J = 7.6 Hz, 2 CH), 7.15 (2 H, d, 3J = 7.5 Hz, 2 CH), 7.24 (2 H, d, 3J = 7.6 Hz, 2 CH), 7.38 (2 H, d, 3J = 7.6 Hz, 2 CH), 7.68 (2 H, d, 3J = 7.5 Hz, 2 CH), 7.82 (1 H, s, CH) ppm. 13C NMR: δ 21.3 (Me), 22.4 (Me), 55.7 (MeO), 92.3 (C), 102.7 (CH), 107.5 (CH), 115.2 (2 CH), 124.2 (2 CH), 125.7 (2 CH), 126.6 (2 CH), 127.3 (2 CH), 129.4 (2 CH), 135.3 (C), 136.4 (C), 137.2 (C), 138.2 (C), 141.4 (C), 157.2 (C), 159.3 (C), 192.3 (C=S) ppm. MS, m/z (%): 429 (M+, 15), 352 (56), 77 (100).

*3-(4-nitrophenyl)-5,6a-bis(4-bromophenyl)-3a,6a-dihydro-furo[2,3-d][1,3]-oxazole-2(3H)-thione* **(4f):**

Yellow powders; mp 187-189 °C, yield: 0.76 g (83%). IR (KBr) (νmax/cm-1): 1698, 1643, 1586, 1467, 1354 and 1295 cm-1. 1H NMR: δ 6.96 (1 H, s, CH), 7.34 (2 H, d, 3J = 7.8 Hz, 2 CH), 7.45 (2 H, d, 3J = 7.7 Hz, 2 CH), 7.52 (2 H, d, 3J = 7.6 Hz, 2 CH), 7.64 (2 H, d, 3J = 7.5 Hz, 2 CH), 7.82 (2 H, d, 3J = 7.8 Hz, 2 CH), 7.86 (1 H, s, CH), 8.04 (2 H, d, 3J = 7.8 Hz, 2 CH), ppm. 13C NMR: δ 93.2 (C), 104.2 (CH), 109.3

(CH), 117.2 (2 CH), 120.4 (C), 123.2 (C), 127.4 (2 CH), 128.2 (2 CH), 129.7 (2 CH), 130.4 (2 CH), 132.2 (2 CH), 135.2 (C), 138.3 (C), 140.2 (C), 147.3 (C), 158.2 (C), 192.5 (C=S) ppm. MS, m/z (%): 574 (M+, 10), 497 (68), 77 (100).

**DPPH radical scavenging test**

**Evaluation of DPPH radical trapping**

By employing of DPPH radical trapping experiment, antioxidant activity of some synthesized compounds **4a-4d** was measured consistent with the reported method by Shimada et al In this experiment, different concentrations of 4a-4d (200-1000 ppm) were added to a same volume of methanolic solution of DPPH (1 mM) and the mixtures were mixed and then put in a dark room. The maximum absorbance of the mixture was 517 nm after 30 min at room temperature. The synthesized compounds **4a-4d** was exchanged with 3 ml methanol in the control sample and butylated hydroxytoluene (BHT) and 2-tert-butylhydroquinone (TBHQ) were used as standard controls. The DPPH performance is calculated by the following formula.

**The power of reducing experiment**

By the procedure of Yildirim et. al. the power of **4a-4d** to reduce iron (III) was measured. The compounds 4a-4d (1 ml) were combined with 2.5 ml of potassium ferricyanide ($K_3Fe(CN)6$; 10 g/L) and 2.5 ml of phosphate buffer (0.2 M, pH 6.6) and stirred for 30 min at 50 °C. Then, 2.5 mL of trichloroacetic acid (10 % w/v) were added to the previous mixture and centrifuged for 10 min. Finally, supernatant (2.5 mL) and 0.5 ml $FeCl_3$ (1 g/L) was combined together in 2.5 ml of distilled water. The absorbance of samples was measured at 700 nm and higher absorbance attributed to higher reducing power.

**Evaluation of antibacterial activity**

The antibacterial effect of synthesized compounds against Gram-positive and Gram-negative bacteria was investigated using the disk diffusion method. All microorganisms were obtained from the Persian type culture collection (PTCC), Tehran, Iran. Microorganisms were cultured for 16 to 24 h at 37°C and prepared to turbidity equivalent to McFarland Standard No. 0.5. Streptomycin and Gentamicin at a concentration 40 $\mu$g/mL, were used as standard against bacteria. The bacterial suspension was prepared to match the turbidity of the 0.5 McFarland (Approximately 1.5 × 108 CFU/mL) standards and cultured with a sterile swab on Mueller Hinton agar. All synthesized compounds were screened for their antibacterial (Gram-positive and Gram-negative) at a concentration of 25 µg/ml that was poured on sterile blank disks. The plates were incubated overnight at 37 °C for 24 h in an incubator. The result was studied by measuring the diameter of the inhibition zone and compared to with the control.

**Acknowledgements**

We gratefully acknowledge from Technical and Vocational University (TVU) of Tehran because of spiritual support.

**References**

[1] Zhu, J.; Bienaymé, H. Multicomponent reactions.Wiley-VCH, Veinheim, **2005**.
[2] Orru, R.V.; Ruijter, E. Synthesis of heterocycles *via* multicomponent reactions II. Springer, Berlin **2010**.
[3]Domling, A.; Wang, W.; Wang, K. *Chem. Rev.* **2012**, *112*, 3083.
[4] Tao, J.; Kazlauskas, R. J. Biocatalysis for Green Chemistry and Chemical Process Development, John Wiley & Sons, **2011**.
[5] (a) Chanda, A. ; Fokin, V.V. *Chem. Rev.* **2009**, *109*, 725.
[6] Butler, R.N.; Coyne, A.G. *Chem. Rev.* **2010**, *110*, 6302.
[7] M.O. Simon, C. Li, *J. Chem. Soc. Rev.* **2012**, *41*, 1415.
[8] S. Chen, F. Ren, S. Niu, X. Liu and Y. J. Che, *Nat. Prod.* ,**2014**, *77*, 9.
[9] Y. Cai, C. C. Ling and D. R. Bundle, *Org. Lett*., **2005**, *7*, 4021; Z. L. Zhan, F. X. Ren and Y. M. Zhao, *Carbohydr. Res.*, **2010,** *345*, 315.
[10] E. Urban and C. R. Noe, *Farmaco*, **2003**, *58*, 243.
[11] J. D. Charrier, S. J. Durrant, J. Studley, L. Lawes and P. Weber, *Bioorg. Med. Chem. Lett.*,**2012**, *22*, 485.
[12] T. M. Meulemans, G. A. Stork, F. Z. Macaev, B. J. M. Jansen and A. de Groot, *J. Org. Chem.*,**1999**, *64*, 9178; D. R. Williams, C. M. Rojas and S. L. Bogen, *J. Org. Chem.*, **1999**, *64*, 736; A. Avenoza, J. H. Busto, C. Cativiela and J. M. Peregrina, *Tetrahedron Lett.*,**2002**, *43*, 4167.
[13] Soleimani-Amiri, S.; Shafaei, F.; Varasteh Moradi, A.; Gholami-Orimi, F.; Rostami, Z. *J. Heterocyclic Chem.*, **2019**, *56*, 2744.
[14] Arisa Ohura, ToshimasaItoh, Hiroaki Ishida, Akio Saito, and Keiko Yamamoto *Asian J. Org. Chem* 10.1002/ajoc.201700074.

[15] Gang Li, Li Li, Haihong Huang and Dali Yin *Org. Biomol. Chem.*,**2015**, *13*, 4418.
[16] Anna, C.G.; Helena, I. M. B.; Scott, G.F.; Clifton, E.B.; Brent, R.C. *Tetrahedron Lett.* **2005,** *46*, 7355.