Available online at www.derpharmachemica.com

Scholars Research Library

Der Pharma Chemica, 2013, 5(3):156-162
(http://derpharmachemica.com/archive.html)

ISSN 0975-413X
CODEN (USA): PCHHAX

1-[2-Substituted ethyl]-2-methyl-5-nitroimidazole derivatives, synthesis and antibacterial activities

Mahmoud A. Al-Sha'er*, Iman Mansi and Nancy Hakooz

Faculty of Pharmacy, Zarqa University, Zarq, Jordan

ABSTRACT

*In this study, seven diverse 1-[2-substituted ethyl]-2-methyl-5-nitroimidazole derivatives were synthesized by reacting various reagents with metronidazole, miscellaneous derivatives were synthesized that represent different scaffolds. The structure elucidation of the compounds was performed by IR, ^{1}H-NMR, ^{13}C-NMR and EI-MS. Antimicrobial activities of the compounds were examined and notable activities were observed. Although metronidazole had no activities against aerobic bacteria, synthetic benzene sulfonated metronidazole derivative (**M1**) exerts some inhibition on Staphylococcus aureus (MIC =250 µg/ml) and phenylacetamide metronidazole derivative (**M3**) exerts certain effect on Streptoccocus B (MIC = 187.5 µg/ml).*

Keywords: metronidazole, phthalic anhydride, gram positive, antimicrobial activity, gram negative.

INTRODUCTION

Some nitroimidazole derivatives (i.e. metronidazole, ornidazole and ronidazole) have become the important agents for a long time for treatment of serious infections caused by anaerobic bacteria and protozoa [1-3]. It has been known that the alcoholic functional group in the metronidazole molecule is suitable for various reactions [4]. Hence it is substituted with different groups by replacing the hydroxide group with several groups lead to effective imidazole derivatives [5-12]. Using these features, as an extension of reported work [13-15] on nitroimidazole, seven compounds 1-[2-substituted- ethyl]-2-methyl-5-nitroimidazoles derived from metronidazole were synthesized and their antibacterial activities were evaluated.

Chemistry

The synthesis of 1-[2-substituted-ethyl]-2-methyl-5-nitroimidazole derivatives were accomplished in the sequence of reactions depicted in figures I and II. To obtain the final products **M1**-**M7**, metronidazole was reacted with a suitable reagents; nucleophilic substitution reaction using the following reagents; benzene sulfonyl chloride (**M1**), phthalic anhydride (**M2**), aniline alkylamide (**M3**), sulfanilamide alkylamide (**M4**), hexylamine alkylamide (**M5**), o-toluidine alkylamide (**M6**) and phenylethylamine alkylamide (**M7**). The structures of the compounds were confirmed by means of spectral measurements. In the Infra Red spectra (IR), the characteristic N=O stretching bands for all compounds were observed at 1519–1526 and 1360–1375 cm^{-1} regions., carbonyl stretching bands appeared as sharp and strong bands in the 1677–1692 cm^{-1} regions. In the NMR spectra, imidazole 2-CH3 protons and 4-H proton resonated as singlets at about 2.6 and 8.0 ppm respectively, imidazole N-Ethyl CH2 protons appeared as triplets at about 3.47–3.85 and 4.17–4.35 ppm. Other protons on the imidazole ring, aliphatic and aromatic protons resonated as expected [15]. In the EI-MS spectra of the compounds fragments were observed instead of molecular

ion peaks. Thus it can be proposed that the *m/z* 46 fragment corresponds to C2H5OH molecule, *m/z* 171 fragment corresponds to metronidazole fragment.

MATERIALS AND METHODS

Reagents and Reference Samples

Pharmaceutical grade metronidazole 99% was supplied by Al-Hikma pharmaceuticals (Amman, Jordan) and certified to contain 99.5%. sulfanilamide (99.5%), aniline (98%), hexylamine(98%), phthalic anhydride, benzene sulfonyl chloride and o-toluidine (99%) , phenylethylamine (95%) and monochloroacetyl chloride (99%) (Sigma-Aldrich, Germany). Bi-distilled water was produced in-house (BOECO Water Still, WS3500, Germany). Isolated pathogenic microbes were supplied from Supporting Medical Sciences Faculty at Zarqa University.

Chemistry

Melting points were measured using Stuart **SMP11** melting point apparatus and are uncorrected. ^{1}H NMR and ^{13}C NMR spectrums were collected on a Varian Oxford NMR-300 spectrometer. Electron Impact mass spectrometry was performed using **ISQ** Thermo Scientific mass spectrometer utilizing electron ionization. Infrared spectra were recorded using Shimadzu **IRPrestige-21** Fourier Transform Infrared spectrophotometer. The samples were analyzed as thin solid films using KBr pellets. Analytical thin layer chromatography (TLC) was carried out using pre-coated aluminum plates and visualized by UV light (at 254 and/or 360 nm).

Synthesis of the *mono*-chloromethyl-acetamide derivatives

To a magnetically-stirred, ice-bathed, solution or suspension of the particular amine (figure II) (1.0 equivalent) and triethylamine (2.0 equivalents) in dry acetone (25 mL), chloroacetylchloride (1.0 equivalent) in dry acetone (25 mL) was gradually added over 30 mins. The reaction mixture was stirred at room temperature until TLC revealed complete consumption of the starting amine. Subsequently, the reaction mixture was poured slowly onto 100 ml of 5% aqueous sodium bicarbonate to neutralize the generated acid. The precipitated crude products were purified by recrystallization from acetone/water [21].

General method for 1-[2-(substituted) ethyl]-2-methyl-5-nitroimidazoles **M1-M7**

M1:Benzenesulfonic acid-2-(2-methyl-5-nitro-imidazole-1-yl)ethyl ester

a mixture of metronidazole (1 mmol, 0.257g), benzene sulfonyl chloride (5 ml) and triethylamine (3 ml) in dioxane (50 mL) was mixed overnight at room temperature. The solvent was evaporated. The residue was treated with water and neutralized with 5% NaHCO3 solution. The precipitate formed was filtered and recrystalized from acetone: water (2:1).

93% yield; m.p. (acetone) 140–141 °C; IR (KBr) ν_{max} (cm^{-1}) 3100, 2920, 1561–1458 (C=N, C=C), 1520, 1364 (N=O); ^{1}H-NMR (300 MHz, DMSO-d6) (ppm): δ = 2.62 (s, 3H), 3.73 (t, *J* = 6.6 Hz, 2H), 4.21 (t, *J* = 6.5 Hz, 2H), 7.40 (d, *J* = 8.8 Hz, 2H), 7.94 (d, *J* = 8.8 Hz, 2H), 7.62 (dd, 1 H), 8.01 (s, 1H). ; ^{13}C-NMR (75 MHz, DMSO-d6) δ = 19.07 (CH3), 34.2 (CH2), 60.2 (CH2), 127.31 (2 x CH), 129.41 (2 x CH),128.56 (CH), 134.57 (C), 141.61 (C), 162.5 (C) ppm; EI-MS: 53.16, 68.12, 77.08, 80.15, 95.16, 123.03, 140.99, 170.06 (100%), 185.00, 204.06, 265.09, 311.06, 312.11 (M+1) ($C_{12}H_{13}N_3O_3S$).

M2: Phthalic acid mono-[2-(2-methyl-5-nitro-imidazole-1-yl)-ethyl] ester

a mixture of metronidazole (1 mmol, 0.257g) and 1mmol of phthalic anhydride was dissolved in 50 ml acetone, reflux at room temperature for 24 hours, Evaporation of acetone lead to colorless needle crystals, recrystalization by acetone : water (2:1) to ensure purity.

72% yield; m.p. (Acetone) 150–151 °C, IR (KBr) ν_{max} (cm^{-1}): 3400 (broad, COOH), 3119,1751(C =O), 1650(C =O),1577–1466 (C=N, C=C), 1521, 1375 (N=O); ^{1}H-NMR (300 MHz, DMSO-d6) (ppm) δ = 2.52 (s, 3H), 3.82 (t, *J* = 5.8 Hz, 2H), 4.43 (t, *J* = 5.7 Hz, 2H), 7.81 (s, 2H), 7.72-8.21(m, 4H), 11.04(s, 1H, OH); ^{13}C-NMR (75 MHz, DMSO-d6) δ = 19.17 (CH3), 34.3 (CH2), 66.4 (CH2), 127.31 (CH), 129.41 (CH),130.56 (CH), 131.3(CH), 133.66 (CH), 134.57 (C), 136.23 (C),141.61 (C), 162.5 (C), 172.34 (C) ppm; EI-MS: 54.05, 69.17, 81.15 (100%), 83.09, 95.11, 105.09, 124.12, 149.07, 171.07, 318.45, 319.48, 320.46 (M+1) ($C_{14}H_{13}N_3O_6$).

***M3* :** 2-[2-(5-Methyl-2-nitro-imidazol-1-yl)-ethoxy]-N-phenyl-acetamide
1mmol, 0.257gm of metronidazole is dissolved in 50 ml dioxane, 2 mmol of Na metal is added , stirred at room temperature for 1 hour, and then filtered, 1mmol of aniline alkyl amide derivative [21] is added, reflux overnight. The whole reaction mixure is neutralized by 150 ml of 5% $NaHCO_3$ solution, Extraction by 50 ml dichloromethane three times.

49% yield; m.p. (acetone) 150-152 °C; IR (KBr) ν_{max} (cm^{-1}): 3100, 2890,1650 (C =O), 1597–1424 (C=N, C=C), 1350 (N=O); ^{1}H-NMR (300 MHz, DMSO-d6) (ppm): δ =2.61 (s, 3H), 3.81 (t, *J* = 7.1 Hz, 2H), 4.01(t, *J* = 7.1 Hz, 2H), 4.35 (s ,1H), 7.08 (t, *J* = 8.2 Hz, 1H), 7.31 (t, *J* =8.2 Hz ,2H), 7.62 (d, *J* =8.2 Hz, 2H), 8.05 (s, 1H) ,10.45 (s, 1H); ^{13}C-NMR (75 MHz, DMSO-d6) δ = 14.68 (CH3), 44.05 (CH2), 48.72 (CH2), 60.2 (CH2), 119.80 (2 x CH), 124.2 (CH), 129.3 (2 x CH), 133.4 (C), 139.0 (C), 152.4 (C), 165.1 (C) ppm, EI-MS : 54.18, 67.28, 81.27, 82.21, 124.21 (100%), 172.24, 296.33, 304.17, 305.12 (M+1) ($C_{14}H_{16}N_4O_4$).

M4: 2-[2-(2-Methyl-5-nitro-imidazol-1-yl)-ethoxy]-N-(4-sulfamoyl-phenyl)-acetamide
1mmol of metronidazole is dissolved in 50 ml dioxane, 2 mmol of Na metal is added , stirred at room temperature for 1 hour, and then filtered, 1mmol of sulfanilamide alkyl amide [21] derivative is added, reflux overnight, The whole reaction mixure is neutralized by 150 ml of saturated NaHCO3, Extraction by 50 ml dichloromethane three times.

56% yield; m.p. (acetone) 170–171 °C; IR (KBr) ν_{max} (cm^{-1}): 3300, 3200, 2900, 1701, (C=O), 1602–1540 (C=N, C=C), 1522,1368 (N=O); ^{1}H-NMR (300 MHz, DMSO-d6) (ppm): δ = 2.56 (s, 3H), 3.38 (s, 1H), 3.67 (t, *J* = 5.1 Hz, 2H), 4.30 (s, 2H), 4.35(t, *J* = 5.0 Hz, 2H), 5.02 (t, *J* = 5.3 Hz, 1H), 7.29 (s, 2H), 7.77 (dd, *J* =5.8, 14.4 Hz, 2H), 8.02 (1H, s); ^{13}C-NMR (75 MHz, DMSO-d6) δ = 14.69 (CH3), 44.01 (CH2), 48.71 (CH2), 60.2 (CH2), 119.43 (2 x CH), 127.26 (2 x CH),133.41(CH), 138.83 (C), 139.39 (C), 141.81 (C), 152.40 (C), 165.65 (C) ppm, EI-MS: 54.13, 64.13, 81.10,92.13, 108.08, 124.13, 156.09, 172.06 (100%), 185.03, 199.06, 232.00, 248.04, 382.28, 383.27, 384.32 (M+1) ($C_{14}H_{17}N_5O_6S$).

M5: N-Hexyl-2-[2-(2-methyl-5-nitro-imidazol-1-yl)-ethoxy]-acetamide
1mmol of metronidazole is dissolved in 50 ml dioxane, 2 mmol of Na metal is added , stirred at room temperature for 1 hour, and then filtered, 1mmol of hexylamine alkyl amide [21] derivative is added, reflux overnight, The whole reaction mixure is neutralized by 150 ml of saturated NaHCO3, Extraction by 50 ml dichloromethane three times.

77% yield; m.p. (acetone) 140-141 °C, IR (KBr) ν_{max} (cm^{-1}): 3211, 2894, 1692 (C=O), 1560–1426 (C=N, C=C), 1519,1366 (N=O); ^{1}H-NMR (300 MHz, DMSO-d6) (ppm): δ = 0.87 (d, *J* = 5.8 Hz, 2H),1.25 (s, 3H), 1.40 (s, 2H), 2.62 (*s*, 3H), 3.07 (d, *J* = 5.8Hz, 2H), 3.37 (d, *J* = 4.8 Hz, 2H), 3.68 (d, *J* = 1.6 Hz, 2H), 4.02 (d, *J* = 4.8 Hz, 2H), 4.36(d, *J* = 4.8 Hz, 2H), 5.02 (t, *J* = 4.8 Hz, 2H), 8.01(d, *J* = 4.3 Hz, 1H); ^{13}C-NMR (75 MHz, DMSO-d6) δ = 14.29 (CH3), 14.65 (CH3), 22.49 (CH2), 26.44 (CH2), 29.26 (CH2), 31.40 (CH2), 39.38 (CH2), 43.12 (CH2), 48.71(CH2), 60.22 (CH2), 133.36 (CH), 138.84 (C), 152.38 (C), 166.16 (C) ppm, EI-MS: 54.16, 67.15, 81.12 (100%), 124.10, 154.0, 172.10, 312.10, 313.09 (M+1) ($C_{12}H_{13}N_5O_5S$).

M6: 2-[2-(2-Methyl-5-nitro-imidazol-1-yl)-ethoxy]-N-o-tolyl-acetamide
1mmol of metronidazole is dissolved in 50 ml dioxane, 2 mmol of Na metal is added , stirred at room temperature for 1 hour, and then filtered, 1mmol of o-toluine alkyl amide derivative [21] is added, reflux overnight, The whole reaction mixure is neutralized by 150 ml of 5% NaHCO3 solution, Extraction by 50 ml dichloromethane three times.
43% yield; m.p. (acetone) 117-118 °C; IR (KBr) ν_{max} (cm^{-1}): 3204, 2960,1689 (C=O), 1597–1424 (C=N, C=C),1521, 1360 (N=O); ^{1}H-NMR (300 MHz, DMSO-d6) (ppm): δ =2.21 (s, 3H), 2.46 (s, 3H), 3.69 (*broad* s, 2H), 4.31 (s, 1H),4.36 (t, *J* = 4.8 Hz, 2H), 5.03 (s,2H), 7.18 (ddd, *J* =6.2, 13.6, 20.2 Hz, 3H), 7.41 (d, *J* =7.5Hz,1H), 8.02 (s, 1H); ^{13}C-NMR (75 MHz, DMSO-d6) δ = 14.66 (CH3), 18.01(CH3), 43.61 (CH2), 48.71(CH2), 60.22 (CH2), 125.46 (CH), 126.14 (CH), 126.50 (CH), 130.83 (CH), 132.42 (C), 133.39 (CH),136.03 (C), 138.85 (C), 153.39 (C), 165.33 (C) ppm , EI-MS: 54.05, 67.09, 81.07 (100%), 91.04, 105.97, 123.99, 133.95, 153.96, 170.96, 182.93, 184.99, 295.98, 317.99, 318.93, 319.97 (M+1) ($C_{15}H_{17}N_5O_5S$).

M7: 2-[2-(2-Methyl-5-nitro-imidazol-1-yl)-ethoxy]-N-phenethyl-acetamide
1mmol of metronidazole is dissolved in 50 ml dioxane, 2 mmol of Na metal is added , stirred at room temperature for 1 hour, and then filtered, 1mmol of phenylethylamine alkyl amide derivative [21] is added, reflux overnight, The whole reaction mixure is neutralized by 150 ml of 5% NaHCO3 solution, Extraction by 50 ml dichloromethane three times.

67% yield; m.p. (acetone) 137-138 °C; IR (KBr) ν_{max} (cm^{-1}): 3214, 2940,1680 (C =O), 1521, 1480,1424,1360 (N=O); ^{1}H-NMR (300 MHz, DMSO-d6) (ppm): δ =2.46 (s, 3H), 2.74 (t, *J* = 7.5Hz, 2H), 3.32 (t, *J* = 7.5Hz, 2H), 3.69 (dd, *J* = 5.2, 10.3 Hz, 2H),4.04 (s, 1H), 4.36 (t, *J* = 5.1Hz, 2H), 5.02 (t, *J* = 5.1Hz, 2H), 7.20-7.31 (m, 5H), 8.02 (s, 1H); ^{13}C-NMR (75 MHz, DMSO-d6) δ = 14.68 (CH3), 35.30 (CH2), 41.0 (CH2), 43.08 (CH2), 48.72 (CH2), 60.2 (CH2), 126.0 (CH), 128.77 (2 x CH),129.08 (2 x CH),133.41 (CH), 138.85(C),139.64(C),152.39(C), 166.28(C) ppm , EI-MS: 333.16 (M+1) ($C_{16}H_{20}N_4O_4$).

Microbiology
Antibacterial activities of the compounds were determined using agar well method to determine zone of inhibition and tube dilution technique for active compounds[22, 23]. The minimum inhibitory concentration (MIC) values are given in µg/mL. The standard bacteria strains used, zone of inhibition and MIC values are shown in table I and table II. Samples were prepared as follows; 0.01 gm dissolved in 1ml of acetone, 25 µl was added in each well using agar well method. 25 µl of acetone was used as blank. Diameter of the inhibition zone was measured. Serial dilution method was used for MIC value determination as follows; 50 µl of the tested compound (0.01 gm/ml) was transferred to 2 ml of nutrient broth, vortex, transfer 1 ml of liquid broth to the next tube, add 1 ml of broth, vortex, then transfer 1 ml of mixed broth to the third test tube, the serial concentrations; 250, 125 and 75 µg/ml.

RESULTS AND DISCUSSION

In one of reported works [16-18] some of the 1-[2-substituted ethyl]-2-methyl-5-nitroimidazole derivatives were highly effective against aerobic bacteria. In analogy, the compounds which were synthesized in this study, also exhibited antibacterial activity as expected (table I, II). They are notably effective against aerobic bacteria, but metronidazole itself is not (MIC > 1024 mg/mL). The highest activity was observed for the compounds **M1** against *Staphylococcus aureus* and **M3** against *Streptococcus B* (MIC = 250, 187.5 µg/ml respectively). The rest compounds exerts variable intermediate effect on *Streptoccocus B* and *Staphylcoccus epidermidis*. However, *Shigella sonnei*, *Pseudomonas aeruginosa* , *Escherichia coli* , *Enteroccocus faecium* and *Proteus mirabilis* which are gram negative bacteria are resistant for all tested synthetic compounds. It is observed that the active synthetic compounds are active on gram positive bacteria but not on gram negative bacteria. The antimicrobial activity of the newly prepared compounds were assessed using antimicrobial susceptibility testing and minimal inhibitory concentration (MIC) values were measured for sensitive compounds (table II) [19]. The spectrum of activity of the tested compounds covered the gram-positive bacteria, but not the gram-negative bacteria, aneorobic bacteria were not tested. The reported antimicrobial activity was lower than that of the reference tested antimicrobial agent; cefdinir [20]. Nevertheless, it was demonstrated that compounds **M1** and **M3** have higher antimicrobial activity towards the *S.aureus* and *Streptoccocus B* respectively than the reference antimicrobial, metronidazole (table II).

Table I. Zone of inhibition of the synthetic compounds (diameter in mm)

Compound	*S.aureus*	*S. epidermidis*	*Shigella sonnei*	*P. mirabilis*	*S. saprophyticus*	*E. faecium*	*Streptococcus B*	*E.coli*	*P. aeruginosa*
M1	**19**	**10**	0	0	**10**	**4**	**8**	**6**	0
M2	**4**	**11**	0	0	**12**	**5**	**12**	**8**	0
M3	**5**	**15**	**10**	**0**	**13**	**10**	**16**	**10**	**10**
M4	**8**	0	0	0	0	**2**	**5**	**12**	0
M5	0	**10**	0	0	**10**	0	**10**	**8**	0
M6	0	**12**	0	0	**16**	0	**15**	0	0
M7	0	**5**	0	0	0	0	0	0	0
metronidazole	0	0	0	0	0	0	**10**	**4**	0
Cefdinir*	**35**	**25**	**16**	**25**	**32**	**38**	**30**	**22**	**8**

**Zone of inhibition is experimental measured values in mm.*

Resistant (1-10), intermediate (11-15), sensitive (> 16) [19].

Table II. Antibacterial activities of the sensitive compounds (MIC µg/mL)

Compound	*S.aureus*	*S. epidermidis*	*Shigella sonnei*	*P. mirabilis*	*S. saprophyticus*	*E. faecium*	*Streptococcus B*	*E.coli*	*P. aeruginosa*
M1	S, MIC 250	R	R	R	R	R	R	R	R
M2	R	I	R	R	I	R	I	R	R
M3	R	I	R	R	I	R	S, MIC= 187.5	R	R
M4	R	R	R	R	R	R	R	I	R
M5	R	R	R	R	R	R	R	R	R
M6	R	I	R	R	I	R	I	R	R
M7	R	R	R	R	R	R	R	R	R
metronidazole	R	R	R	R	R	R	R	R	R
Cefdinir* MIC µg/ml	S 0.5	S 0.5	S 0.25	S 0.125	S 0.5	S 8	S 0.06	S 0.12	R

MIC values reported [20]

Resistance I, Intermediate (I), Sensitive (S) [19].

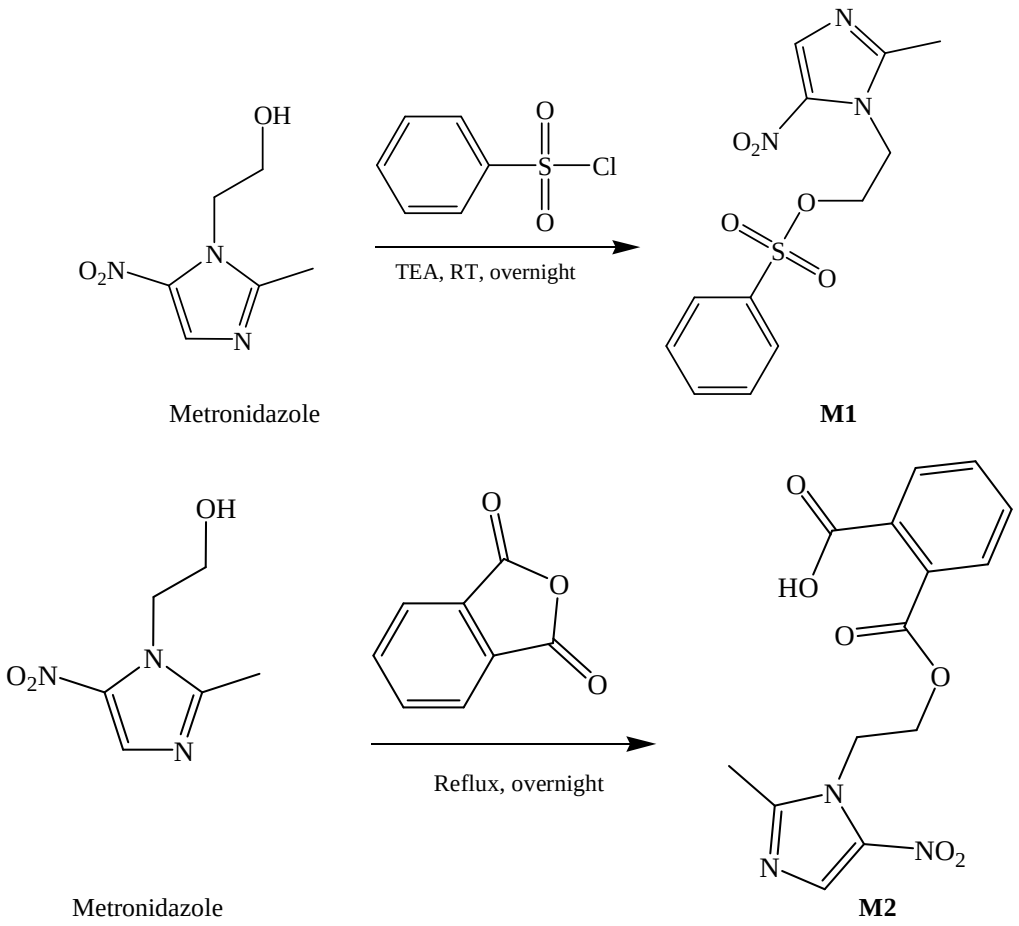
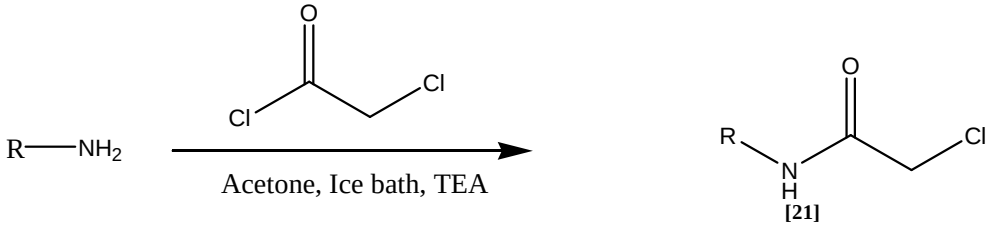
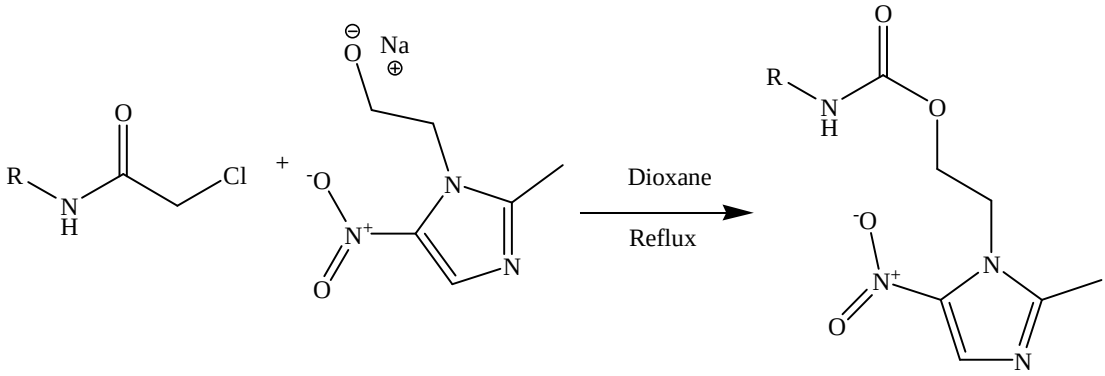


Figure I: Scheme I for synthetic compounds M1 and M2.



[21]



M3-M7

Compound	Amine used	R
M3	Aniline	
M4	Sulfanilamide	
M5	Hexylamine	
M6	o-Toluidine	
M7	Phenylethylamine	

Figure II: Scheme II for synthetic compounds M3 – M7

CONCLUSION

The synthetic metronidazole derivatives **M1** and **M3** had a narrow spectrum of activity (MIC = 250, 187.5 µg/ml) against *S.aureus* and *Streptoccocus B* respectively. Nevertheless they are superior than the metronidazole itself. The alkylamide ether is considered as a new scaffold with antimicrobial activity against gram positive bacteria, further study of the structural activity relationship could guide the modifications toward more potential agents.

Acknowledgment
The authors wish to acknowledge the deanship of Scientific Research at Zarqa University for supporting this work. Many thanks for Enas Radwan, Shereen Al-Khateeb and Sahar Edaily for measuring the melting points, TLC analysis and MS analysis.

REFERENCES

[1] DI Edwards, *Antibiot Chemother*, **1993**, 3, 19-20.
[2]G Navarrete-Va´zquez, MM Rojano-Vilchis, L. Ye´ pez-Mulia, V. Mele´ ndez, L. Gerena, A. Herna´ndez-Campos, R. Castillo, F. Herna´ ndez-Luis, *Eur J Med Chem*, **2006**, 41, 135–141.
[3] M Boiani, M Gonza´ lez, *Mini Rev Med Chem*, **2005**, 5, 409–424.
[4] BM Mallia, A Mathur, S Subramanian, S Banerjee, HD Sarma, M Venkatesh, *Bioorg Med Chem Lett*, **2005**, 15, 3398-3401.
[5] A. Buschini, F. Giordani, CN de Albuquerque, C Pellacani, G Pelosi, C Rossi, TM Zucchi, P Poli, *Biochem Pharmacol*, **2007**, 73, 1537–1547.
[6] MD Crozet, CL Botta, M Gasquet, C Curti, VR Musat, SB Hutter, O Chapelle, N Azas, MD Meo, P Vanelle, *Eur J Med Chem*, **2009**, 44, 653-659
[7] K Walczak, A Gondela, J Suwin´ ski, *Eur J Med Chem*, **2004**, 39, 849–853.
[8] N Hadj-esfandiari, L Navidpour, H Shadnia, M Amini, N Samadi, MA Faramarzi, A Shafiee, *Bioorg Med Chem Lett*, **2007**, 17, 6354–6363.
[9] S Khabnadideh, Z Rezaei, A Khalafi-Nezhad, R Bahrinajafi, R Mohamadi, AA Farrokhroz, *Bioorg Med Chem Lett*, **2003**, 13, 2863–2865.
[10] D Olender, JZ_wawiak, V Lukianchuk, R Lesyk, A Kropacz, A Fojutowski, L Zaprutko, *Eur J Med Chem*, **2009**, 44, 645-652.
[11] MB Mallia, S Subramanian, S Banerjee, HD Sarma, M Venkatesh, *Bioorg Med Chem*, **2006**, 14, 7666–7670.
[12] R Miri, KA Javidnia, H Sarkarzadeh, B Hemmateenejad, *Bioorg Med Chem*, **2006**, 14, 4842–4849.
[13] Ş Demirayak, N Kiraz, *Farmaco*, **1993**, 48, 443-446.
[14] M Wen-Jun, L Peng-Cheng, S Lei, L Huan-Qiu, Z Hai-Liang, *Bioorg Med Chem* , **2009**, 17, 7531–7536.
[15] AK Atia, *Molecules*, **2009**, 14, 2431-2446.
[16] HA Saadeh, IM Mosleh, AG Al-Bakri , MS Mubarak, *Monatsh Chem*, **2010**, 141, 471–478.
[17] E Herna´ndez-Nu´ n˜ez, H Tlahuext , R Moo-Puc , H Torres-Go´mez , R Reyes-Martı´nez , R Cedillo-Rivera , C Nava-Zuazo , G Navarrete-Vazquez, *Eur J Med Chem*, **2009,** 44, 2975–2984.
[18] S Demirayaka, AÇ Karaburun, N Kiraz, *Eur J Med Chem*, **1999**, 34, 275−278.
[19] A guide to sensitivity testing: Report of working party on antibiotic sensitivity testing of the British Society for Antimicrobial Chemotherapy. *J Antimicrob Chemothrap*, **1991**; 27: Supplement D, P. 1-50.
[20] MA Cohen, ET Joannides, GE Roland, MA Meservey, MD Huband, MA Shapiro, J C Sesnie, CL Heifetz, *Diagn Micr Infec Dis*, **1994**, 18, 31–39.
[21] MA Al-Sha'er, MO Taha, *Med Chem Research*, **2011**, 21, 487-510.
[22]MR McGinnis, MG Rinaldi, In: Lorian V. (Ed.), Antibiotics in Laboratory Medicine, 3rd ed., Williams and Wilkins, London, **1991**, p. 198-199.
[23] M Rodetsky, RC Wheeler, MH Roe, JK Todd, *J Clin Microbiol*, **1986**, 24, 600-606.