Erzincan Üniversitesi
Fen Bilimleri Enstitüsü Dergisi
2017, 10(2), 255-265
Araştırma Makalesi

Erzincan University
Journal of Science and Technology
2017, 10(2), 255-265
Research Article

Synthesis, Characterization and Antimicrobial Activity of Novel Etoksibenzilidenamino Eerived 1,2,4-triazoles

Faruk KARDAŞ[1]*, Murat BEYTUR[2], Haydar YÜKSEK[2], Engin KILIÇ[3], Önder ALBAYRAK[4]

[1]Erzincan University, Education Faculty, 24100 Erzincan, Turkey

[2]Kafkas University, Department of Chemistry, 36100 Kars, Turkey

[3]Erzincan University, Faculty of Pharmacy, 24100 Erzincan, Turkey

[4]Kafkas University, Education Faculty, 36100 Kars, Turkey

(Geliş Tarihi/Received:20/07/2017, Kabul Tarihi/Accepted: 12/10/2017)

ABSTRACT

The benzaldehyde and their derivatives have a good antimicrobial properties. Our research aim is to improve the antimicrobial effect, different derived from *3-Alkil/aril*-4 triozole and metoksibenzoksi. The synthesized triazoles were characterized by spectroscopic techniques, such as, ^{1}H NMR, ^{13}C NMR, UV absorption spectra, and mass spectra (MS). We were mesured their antibacterial activities in vitro against *Bacillius subtilis, Yersinia enterocolitca, Bacillus cereus, Staphlacoccus aureus, Esherichia coli, Pasterulla multicida, Klepsiella pnemonias*. The synthesis of a series of etoksibenzilidenamino derived 1,2,4-triazoles has been realized with good yields using the CuAAC of a variety of alkynyl heterocycles, alkynyl carbohydrates or alkynes. The structures of the obtained compounds were confirmed by NMR spectroscopy (^{1}H and ^{13}C) and mass spectrometry. Overall, the molecules are not showed very antibacterial activity, but our antibiotic showed that important antibacterial activity.

Keywords: *3-Alkil/aril*-1,2,4-triazole, benzaldehyde, Antibacterial activity, Disc diffusion method

1,2,4-triyazolden Kaynaklanan Yeni Kuşak Etoksibenzilidenaminonun Sentez, Karekterizasyon ve Antimikrobiyel Akvitesi

ÖZ

Benzaldehit ve türevleri iyi bir antimikrobiyal özelliklere sahiptir. Bu Çalışmanın amacı, 3-Alkil / aril-4 triozole ve metoksibenzoksi'den elde edilen farklı antimikrobiyal etkinin geliştirilmesidir. Sentezlenen triazoller, 1H NMR, ^{13}C NMR, UV absorpsiyon spektrumu ve kütle spektrumu (MS) gibi spektroskopik teknikler ile karakterize edildi. Antibakteriyel faaliyetlerini *Bacillius subtilis, Yersinia enterocolitca*, *Bacillus cereus, Staphlacoccus aureus*, *Esherichia coli, Pasterulla multicida, Klepsiella pneumoniae*'lere karşı in vitro olarak uygulandı.. Etoksibenzilidenamino türevi 1,2,4-triazollerin bir sentezi, çeşitli alkinil heterosikllerin, alkinil karbonhidratların veya alkinlerin CuAAC kullanılarak iyi verim ile gerçekleştirildi. Elde edilen bileşiklerin yapıları NMR spektroskopisi (1H ve 13C) ve kütle spektrometrisi ile teyit edildi. Genel olarak, moleküller çok antibakteriyel aktivite göstermedi; ancak antibiyotik önemli antibakteriyel aktivite gösterdi.

Anahtar Kelimeler: 3-Alkil/aril-1,2,4-triazole, benzaldehyde, Antibacterial etki, Disc difuzyon yöntemi

ISSN: 1307-9085
e-ISSN: 2149-4584

*Correspondence to:fkardas2400@gmail.com

DOI: 10.18185/erzifbed.329597

1. Introduction

Recently, the rates of microbial threats associated with the increasing emergence of antimicrobial resistance in hospitals are major concerns for public health around the world (Fichtali W., et al., 2016). Especially numerous multi-drug resistant Gram-positive bacteria pathogens, which are methicillin-resistant like Staphylococcus aureus, are growing threat to human health, like others pathogenic bacteria (Fichtali W., et al., 2016). This has required new efforts for the development of new powerful antimicrobial agents with broad spectrum of activity that has an important role to control the emerging multi-drug resistance strains of bacteria (Chu T., et al., 1996 and Chua D.T., et al., 2008).

The benzophenones (still called diphenyl ketones) are a class of molecules obtained by two pathways, natural and synthetic, and are pharmacologically active compounds (Karrer, et al., 2000 and Henry et al, 1999). The 1,2,3-triazoles and their derivatives which have become a class of very active compounds with a broad spectrum of chemotherapeutic activities (Thirumurugan P., etal., 2013 and Kharb R., et al., 2011). Many 1,2,3-triazoles, are proved potent and having several biological properties, such as, antiviral [He Y., et al., 2014], antiepileptic [Ulloora S., et al., 2013], antifungal [Darandale S.N. et al. 20123), antibacterial [Kamal A. et al., 213], antimicrobial activities [Kaushik C.P., et al., 2014], [27]. These 1,2,3-triazoles are incorporated in medicinal chemistry and form some drugs available in markets. Among the most well-known structures (Figure 1), we cite 3-Alkil/aril-4-[3-etoksi-2-(4-metoksibenzoksi)-benzilidenamino]-4,5-dihidro-1H-1,2,4-triazol-5-one.

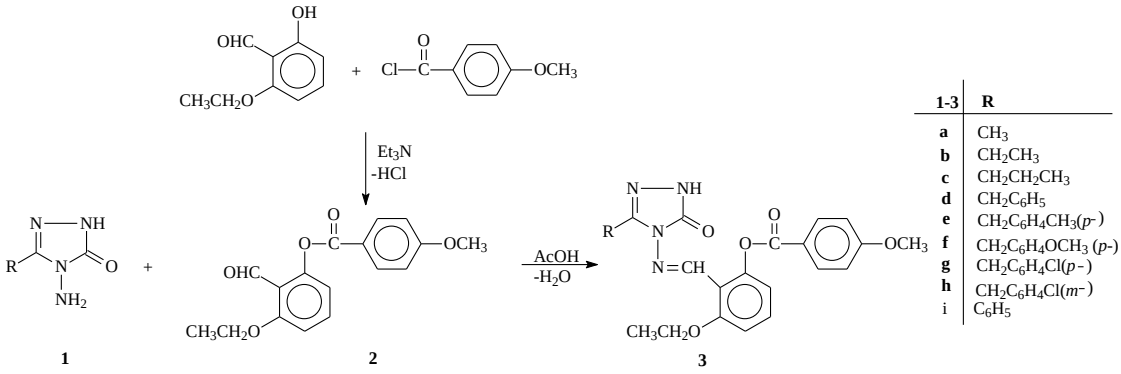

Figure1: Synthesis route of compounds 2, 3

Based on all the previous informations, and in the interest of synthesis of new antimicrobial agents, our research aims the incorporation of the benzaldehyde, because both moieties are known for their antimicrobial properties (Tarık, E.S.A., 2009). In order to improve the antimicrobial effect, different derived from 3-Alkil/aril-4 triozole and metoksibenzoksi. The synthesized triazoles were characterized by spectroscopic techniques, such as, 1H NMR, 13C NMR, UV absorption spectra, and mass spectra (MS). We were evaluated for their antibacterial activities in vitro against

Bacillius subtilis, Yersinia enterocolitca, Bacillus cereus, Staphlacoccus aureus, Esherichia coli, Pasterulla multicida, Klepsiella pnemonias.

2. Material and Methods

2.1. General procedure for the synthesis of compound

Chemical reagents and all solvents used in this study were purchased from Merck AG, Aldrich and Fluka. Melting point was determined in open glass capillary using a WRS-2A Microprocessor Melting point apparatus and is uncorrected. The IR spectra were obtained on an ALPHA-P BRUKER FT-IR spectrometer. 1H and 13C NMR spectra were recorded in deuterated dimethyl sulfoxide with TMS as internal standard using a Varian Mercury spectrometer at 400 MHz and 100 MHz, respectively. UV absorption spectra were measured in 10 mm quartz cells between 200 and 400 nm using a Schimadzu-1201 UV-Visible spectrometer. Extinction coefficients (□) are expressed in L mol-1 cm-1. Elemental analysis of the new compound was made in Leco 932 Elemental Combustion System CHNS-O elemental analyser.

General procedure for the synthesis of compound 35(a)- 3-Alkil/aril-4-[3-etoksi-2-(4-metoksibenzoksi)-benzilidenamino]-4,5-dihidro-1H-1,2,4-triazol-5-on:

3-Methyl-4-amino-4,5-dihydro-1H-1,2,4-triazol-5-one (3a) (1.14g, 0.01 mol) was dissolved in acetic acid (30 mL) and treated with 3-etoksi-2-(4-metoksibenzoksi) benzaldehit (35) (3.00g, 0.01 molThe mixture was refluxed for 2 h and then evaporated at 50-55 °C in vacuo) .Several recrystallizations of the residue from acetic acid-water gave pure compound 3-Metil-4-[3-etoksi-2-(4-metoksibenzoksi)-benzilidenamino]-4,5-dihidro-1H-1,2,4-triazol-5-on (37) as colorless crystals).

Yield: 3.51 g (88.65 %); mp: 205 °C; IR (KBr, υ, cm−1): 3183 (NH), 1734, 1697 (C=O), 1602 (C=N), 1258 (COO), 833 (1,4-disubstituted aromatik ring) ; 1H NMR (400 MHz, DMSO-d6): δ 1.30 (t, 3H, OCH2CH3, J=7.00 Hz); 2.22 (s, 3H, CH3), 3.98 (s, 3H,OCH3), 4.07(q, 2H, OCH2, J=7.00 Hz); 7.00(d, 2H, J=8.92 Hz); 7.08(d, H, J=8.16 Hz);7.25-7.29(m,H);7.56(d,H, J=7.96 Hz); 8.19(d,2H, J=7.05 Hz); 9.95 (s, 1H, N=CH), 9.18 (s, 1H, NH); 13C NMR (100 MHz, DMSO-d6): δ 11.45 (CH3); 14.71(2OCH2CH3);55.18(OCH3); 64.83(OCH2CH3);113.96;116.05;118.76;121.32;126.48;127.70(2C);132.53;140.54;150.49; 163.99(2C), (arom-C), 145.89 (triazole C3), 150.49 (N=CH), 151.41 (triazole C5), 164.23 (COO); UV λ_{max} (ε): 254 (25708), 228 (23308), 218 (23408) nm.

General procedure for the synthesis of compound 38 (b)- 3-Etil--4-[3-etoksi-2-(4-metoksibenzoksi)-benzilidenamino]-4,5-dihidro-1H-1,2,4-triazol-5-on (38):

3-Ethyl-4-amino-4,5-dihydro-1H-1,2,4-triazol-5-one (3b) (1.28g, 0.01 mol) was dissolved in acetic acid (30 mL) and treated with 3-etoksi-2-(4-metoksibenzoksi) benzaldehit (35) (3.00g, 0.01 mol) 1. The mixture was refluxed for 2 h and then evaporated at 50-55 °C in vacuo. Several

recrystallizations of the residue from acetic acid-water gave pure compound 3-Etil-4-[3-etoksi-2-(4-metoksibenzoksi)-benzilidenamino]-4,5-dihidro-1H-1,2,4-triazol-5-on (38) as colorless crystals.

Yield: 3.37 g (82.27 %); mp: 178 ºC; IR (KBr, υ, cm−1): 3162 (NH), 1738, 1697 (C=O), 1603 (C=N), 1254 (COO), 842 (1,4-disubstituted aromatik ring) ; 1H NMR (400 MHz, DMSO-d6): δ 1.29 (t, 3H, OCH2CH3, J=6.96 Hz); 1.20 (t, 3H, CH3, J=7.48 Hz); 2.59(q, 2H, CH2, J=7.52 Hz);3.86 (s, 3H,OCH3), 4.06(q, 2H, OCH2, J=7.00 Hz); 7.99(d, 2H, J=8.96 Hz); 7.07(d, H, J=8.24 Hz); 7.26 (t, H, J=8.04 Hz); 7.54 (d,H, J=8.00 Hz); 8.20(d,2H, J=6.92 Hz); 9.96 (s, 1H, N=CH); 10.2 (s, 1H, NH); 13C NMR (100 MHz, DMSO-d6): δ 10.14 (CH2CH3); 14.71(2OCH2CH3);19.06(CH2CH3);55.51(OCH3);64.83(OCH2CH3);113.96;116.00;118.81;121.34;126.47;127.77(2C);132.54;140.42;151.42;163.99(2C), (arom-C); 149.54 (triazole C3), ⌈150.41 (N=CH), 152.14 (triazole C5), 164.24 (COO); UV λ_{max} (ε): 260 (23630), 228 (20937), nm; Anal. Calcd. for C21 H22N4O5 (410.43): C, 61.46; H, 5.40; N, 13.65. Found: C, 61.28; H, 5.27; N, 13.52.

General procedure for the synthesis of compound 39 (c) - 3-n-Propil-4-[3-etoksi-2-(4-metoksibenzoksi)-benzilidenamino]-4,5-dihidro-1H-1,2,4-triazol-5-on (39):

3-n-propil -4-amino-4,5-dihydro-1H-1,2,4-triazol-5-one (3c) (1.42g, 0.01 mol) was dissolved in acetic acid (30 mL) and treated with 3-etoksi-2-(4-metoksibenzoksi) benzaldehit (35) (3.00g, 0.01 mol) 1. The mixture was refluxed for 2 h and then evaporated at 50-55 °C in vacuo. Several recrystallizations of the residue from acetic acid-water gave pure compound 3-n-Propil-4-[3-etoksi-2-(4-metoksibenzoksi)-benzilidenamino]-4,5-dihidro-1H-1,2,4-triazol-5-on (39) as colorless crystals.

Yield: 4.06 g (95.80 %); mp: 177 ºC; IR (KBr, υ, cm−1): 3176 (NH), 1738, 1697 (C=O), 1603 (C=N), 1252(COO), 845 (1,4-disubstituted aromatik ring) ; 1H NMR (400 MHz, DMSO-d6): δ 1.29 (t, 3H, OCH2CH3, J=6.96 Hz); 0.94 (t, 3H, CH3, J=7.44 Hz); 1.68 (sext, 2H, CH2, J=7.44 Hz); 3.87(s,3H,OCH3); 2.57 (t, 2H, CH2, J=7.74 Hz); 4.06(q, 2H, OCH2,

J=7.00 Hz); 7.00(d, 2H, J=8.88 Hz); 7.07(d, H, J=8.16 Hz); 7.27 (t, H, J=8.00 Hz); 7.55 (d,H, J=7.96 Hz); 8.20(d,2H, J=7.12 Hz); 9.96 (s, 1H, N=CH); 10.07 (s, 1H, NH); 13C NMR (100MHz,DMSO-d6):δ13.65(CH2CH2CH3);14.71(OCH2CH3); 19.29(CH2CH2CH3);27.26(CH2CH2CH3);55.51(OCH3);64.83(OCH2CH3);113.98;116.00;118.66;121.32;126.47;127.79(2C);132.54;140.59;151.41;164.00(2C),(arom-C);149.54 (triazole C3);150.36 (N=CH), 152.06 (triazole C5), 164.24 (COO); UV λ_{max} (ε): 260 (30138), 218 (26766), nm.

3-Alkyl(Aryl)-4-[3-ethoxy-2-(4-methoxybenzoxy)-benzylidenamino]-4,5-dihidro-1H-1,2,4-triazol-5-ones (3a-i):

3-Benzil -4-amino-4,5-dihydro-1H-1,2,4-triazol-5-one (3d) (1.90g, 0.02 mol) was dissolved in acetic acid (30 mL) and treated with 3-etoksi-2-(4-metoksibenzoksi) benzaldehit (35) (3.00g, 0.01 mol) 1. The

mixture was refluxed for 2 h and then evaporated at 50-55 °C in vacuo. Several recrystallizations of the residue from acetic acid-water gave pure compound 3-Benzil -4-[3-etoksi-2-(4-metoksibenzoksi)-benzilidenamino]-4,5-dihidro-1H-1,2,4-triazol-5-on (38) as colorless crystals.

Yield: 4.39 g (92.94 %); mp: 165 °C; IR (KBr, υ, cm−1): 3160 (NH), 1742, 1701 (C=O), 1603 (C=N), 1254 (COO), 841 (1,4-disubstituted arom ring) ; 765-699(monosubstitued arom ring);1H NMR (400 MHz, DMSO-d6): δ 1.29 (t, 3H, OCH2CH3, J=6.96 Hz); 3.83(s,3H,OCH3); 3.94(s,2H,CH2); 4.06(q, 2H, OCH2, J=6.96 Hz); 6.97(d, 2H, J=6.96 Hz); 7.08(d, H, J=8.24 Hz); 7.23-7.32 (m,6H,arom ring); 7.51 (d,H, J=7.96 Hz); 8.18(d,2H, J=6.96 Hz); 9.93 (s, 1H, N=CH); 9.66 (s,1H,NH); 13CNMR(100MHz,DMSO-d6):δ 14.71(2OCH2CH3); 31.71(CH2Ph); 55.49(OCH3); 64.83(OCH2CH3); 113.99;116.05;118.46;121.24;126.50;127.13;127.70(2C);128.65(2C);129.06(2C);132.53;134.92;140.69;151.40;164.01(2C), (arom-C); 147.63 (triazole C3), 150.28 (N=CH), 151.76 (triazole C5), 164.21 (COO); UV λ_{max} (ε): 260 (28960), 218 (26430),212(24730) nm; Anal. Calcd. for C26 H24N4O5 (472.50) C, 66.09; H, 5.12 ; N, 11.86. Found: C, 66.00; H, 5.25; N, 11.75.

General procedure for the synthesis of compound 41 (e) - 3-p-Metilbenzil-4-[3-etoksi-2-(4-metoksifenilkarboniloksi)-benziliden-amino]-4,5-dihidro-1H-1,2,4-triazol-5-on (41): 3-p-Metilbenzil -4-amino-4,5-dihydro-1H-1,2,4-triazol-5-one (3e) (2.04g, 0.01 mol) was dissolved in acetic acid (30 mL) and treated with 3-etoksi-2-(4-metoksifenilkarboniloksi) benzaldehit (35) (3.00g, 0.01 mol) 1. The mixture was refluxed for 2 h and then evaporated at 50-55 °C in vacuo. Several recrystallizations of the residue from acetic acid-water gave pure compound 3-p-Metilbenzil -4-[3-etoksi-2-(4-metoksifenilkarboniloksi)-benzilidenamino]-4,5-dihidro-1H-1,2,4-triazol-5-on (41) as colorless crystals.

Yield: 4.55 g (93.54 %); mp: 170 °C; IR (KBr, υ, cm−1): 3182 (NH), 1739, 1701 (C=O), 1600 (C=N), 1256 (COO), 846 (1,4-disubstituted arom ring) ; 1H NMR (400 MHz, DMSO-d6): δ 1.29 (t, 3H, OCH2CH3, J=6.96 Hz); 2.30(s,3H,CH3); 2.84 (s, 3H,OCH3); 3.90 (s, 2H,CH2); 4.06(q, 2H, OCH2, J=6.96 Hz); 6.97(d, 2H, J=7.08 Hz); 7.07-7.17(m,5H, arom ring); 7.25-7.29(m,H, arom ring); 7.53(d, H, J=7.96 Hz);8.18 (d,2H, J=6.96 Hz); 9.93 (s, 1H, N=CH); 9.60(s,1H,NH); 13CNMR (100MHz,DMSO-d6):δ 14.71(OCH2CH3); 21.10(PhCH3); 31.31(CH2Ph); 55.49(OCH3); 64.84(OCH2CH3); 113.99; 116.03; 118.46; 121.25; 126.48; 127.73(2C); 128.99(2C); 129.34; 131.76(2C); 132.53; 136.74; 140.70; 151.40; 164.00 (2C),(arom-C); 147.63 (triazole C3), 150.23 (N=CH), 151.76 (triazole C5), 164.21 (COO); UV λ_{max} (ε): 260 (34536), 224 (31390), nm; Anal. Calcd. for C26 H27N4O5 (486.53): C, 66.66; H, 5.39; N, 11.52. Found: C, 66.85; H, 5.50; N, 11.39.

General procedure for the synthesis of compound 42 (f) - 3-p-Metoksibenzil-4-[3-etoksi-2-(4-metoksibenzoksi)-benzilidenamino]-4,5-dihidro-1H-1,2,4-

triazol-5-on (42): 3-p-Metoksibenzil -4-amino-4,5-dihydro-1H-1,2,4-triazol-5-one (3g) (2.20g, 0.01 mol) was dissolved in acetic acid (30 mL) and treated with 3-etoksi-2-(4-metoksibenzoksi) benzaldehit (35) (3.00g, 0.01 mol) 1. The mixture was refluxed for 2 h and then evaporated at 50-55 °C in vacuo. Several recrystallizations of the residue from acetic acid-water gave pure compound 3-p-Metoksibenzil -4-[3-etoksi-2-(4-metoksibenzoksi)-benzilidenamino]-4,5-dihidro-1H-1,2,4-triazol-5-on (42) as colorless crystals

Yield: 4.73 g (94.20 %); mp: 169 ºC; IR (KBr, υ, cm−1): 3176 (NH), 1741, 1700 (C=O), 1604 (C=N), 1255 (COO), 845 (1,4-disubstituted aromatik ring) ; 1H NMR (400 MHz, DMSO-d6): δ 1.29 (t, 3H, OCH2CH3, J=7.00 Hz); 3.77(s,3H,CH3); 3.84 (s, 3H,OCH3); 3.87 (s, 2H,CH2); 4.07(q, 2H, OCH2, J=7.00 Hz); 6.83(d, 2H, J=6.78 Hz); 6.97(d, 2H, J=6.96 Hz); 7.08(d, 2H, J=8.24 Hz); 7.19(d, 2H, J=6.80 Hz); 7.25-7.29(m,H, arom ring); 7.53(d,H, J=7.96 Hz); 8.18(d, 2H, J=6.92 Hz); 9.93 (s, 1H, N=CH); 9.61 (s, 1H, NH); 13C NMR (100 MHz, DMSO-d6): δ 14.71(OCH2CH3);30.86(CH2Ph); 55.27(OCH3);64.84(OCH2CH3); 113.98(2C); 114.07(2C); 118.51; 121.25; 126.50;126.84;127.73(2C);130.13(2C);132.53(2C);140.68;151.41;151.75;158.67;164.01(2C), (arom-C); 147.97 (triazole C3), 150.31 (N=CH), 151.75 (triazole C5), 164.21(COO); UV λ_{max} (ε): 260 (30138), 218 (26766), nm.

General procedure for the synthesis of compound 43 (g) - 3-p-Klorobenzil-4-[3-etoksi-2-(4-metoksibenzoksi)-benzilidenamino]-4,5-dihidro-1H-1,2,4-triazol-5-on (43): 3-p-klorobenzil -4-amino-4,5-dihydro-1H-1,2,4-triazol-5-one (3İ) (2.245g, 0.01 mol) was dissolved in acetic acid (30 mL) and treated with 3-etoksi-2-(4-metoksibenzoksi) benzaldehit (35) (3.00g, 0.01 mol) 1. The mixture was refluxed for 2 h and then evaporated at 50-55 °C in vacuo. Several recrystallizations of the residue from acetic acid-water gave pure compound 3-3-p-klorobenzil -4-[3-etoksi-2-(4-metoksibenzoksi)-benzilidenamino]-4,5-dihidro-1H-1,2,4-triazol-5-on (43) as colorless crystals.

Yield: 5.03 g (99.31 %); mp: 180 ºC; IR (KBr, υ, cm−1): 3173 (NH), 1740, 1701 (C=O), 1603 (C=N), 1255 (COO), 846-806 (1,4-disubstituted aromatik ring) ; 1H NMR (400 MHz, DMSO-d6): δ 1.29 (t, 3H, OCH2CH3, J=6.96 Hz); 3.83(s,3H,OCH3); 3.89 (s, 2H,CH2); 4.07(q, 2H, OCH2, J=6.96 Hz); 6.96(d, 2H, J=6.96 Hz); 7.09(d, H, J=8.20 Hz); 7.18-7.29(m,5H.arom H);7.47(d, H, J=7.96 Hz);8.17(d, 2H, J=6.92 Hz); 9.91 (s, 1H,N=CH);9.78 (s,H,NH);13CNMR(100MHz,DMSO-d6):δ 14.70(OCH2CH3);31.05(CH2Ph);55.49(OCH3);64.84(OCH2CH3);113.98(2C); 116.15(2C); 118.58; 121.22;126.54;127.57(2C);128.79(2C);130.43(2C);132.53;133.06;133.34;140.65;151.45;164.02(2C), (arom-C); 147.14 (triazole C3), 150.70 (N=CH), 151.73 (triazole C5),

164.21(COO); UV λ_{max} (ε): 258 (34911), 226 (32975), nm.

General procedure for the synthesis of compound 44 (h) - 3-m-Klorobenzil-4-[3-etoksi-2-(4-metoksibenzoksi)-benzilidenamino]-4,5-dihidro-1H-1,2,4-triazol-5-on (44): 3-m-klorobenzil -4-amino-4,5-dihydro-1H-1,2,4-triazol-5-one (3h) (2.245g, 0.01 mol) was dissolved in acetic acid (30 mL) and treated with 3-etoksi-2-(4-metoksibenzoksi) benzaldehit (35) (3.00g, 0.01 mol) 1. The mixture was refluxed for 2 h and then evaporated at 50-55 °C in vacuo. Several recrystallizations of the residue from acetic acid-water gave pure compound 3-3-m-klorobenzil -4-[3-etoksi-2-(4-metoksibenzoksi)-benzilidenamino]-4,5-dihidro-1H-1,2,4-triazol-5-on (44) as colorless crystals.

Yield: 4.83 g (95.38 %); mp: 184 ºC; IR (KBr, υ, cm−1): 3179 (NH), 1737, 1697 (C=O), 1604 (C=N), 1255 (COO), 844-810 (1,4-disubstituted arom ring) ; 1H NMR (400 MHz, DMSO-d6): δ 1.29 (t, 3H, OCH2CH3, J=6.96 Hz); 3.84 (s,3H,OCH3); 3.89 (s, 2H,CH2); 4.06(q, 2H, OCH2, J=6.96 Hz); 6.96(d, 2H, J=6.96 Hz arom ring); 7.07(d, H, J=8.21 Hz); 7.13-7.15(m, H.arom H);7.21-7.29(m, H.arom H);7.46 (d, H, J=8.00 Hz);8.17(d, 2H, J=6.92 Hz); 9.92(s, 1H, N=CH);9.90(s,H,NH);13CNMR(100MHz,DMSO-d6):δ14.70(OCH2CH3); 31.39(CH2Ph);55.49(OCH3); 64.84(OCH2CH3); 113.99; 116.15; 118.63; 121.22;126.57;127.22;127.37;127.57; 129; 35; 129.87(2C); 132.53; 134; 36; 136.85; 140.64; 151.42; 164.02(2C), (arom-C); 146.85 (triazole C3), 150.59 (N=CH), 151.78 (triazole C5), 164.21(COO); UV λ_{max} (ε): 260 (32038), 218 (31607), nm.

General procedure for the synthesis of compound 45 (i) - 3-Fenil-4-[3-etoksi-2-(4-metoksibenzoksi)-benzilidenamino]-4,5-dihidro-1H-1,2,4-triazol-5-on (45):

3-Fenil-klorobenzil -4-amino-4,5-dihydro-1H-1,2,4-triazol-5-one (3İ) (1.76g, 0.01 mol) was dissolved in acetic acid (30 mL) and treated with 3-etoksi-2-(4-metoksibenzoksi) benzaldehit (35) (3.00g, 0.01 mol) 1. The mixture was refluxed for 2 h and then evaporated at 50-55 °C in vacuo. Several recrystallizations of the residue from acetic acid-water gave pure compound 3-3-Fenil-klorobenzil -4-[3-etoksi-2-(4-metoksibenzoksi)-benzilidenamino]-4,5-dihidro-1H-1,2,4-triazol-5-on (45) as colorless crystals.

Yield: 2.64 g (57.57 %); mp: 206 ºC; IR (KBr, υ, cm−1): 3156 (NH), 1744, 1692 (C=O), 1601 (C=N), 1251 (COO), 843 (1,4-disubstituted aromatik ring) ; 1H NMR (400 MHz, DMSO-d6): δ 1.27 (t, 3H, OCH2CH3, J=6.96 Hz); 3.79(s,3H,OCH3); 4.06(q, 2H, OCH2, J=6.96 Hz); 6.98(d, 2H, J=8.88 Hz); 7.07(d, H, J=8.16 Hz); 7.24(t, H, J=8.08 Hz); 7.43-7.46(m,3H.arom H); 7.56(d, H, J=7.92 Hz); 7.90-7.93(m,2H,arom ring); 8.18(d,2H,J=7.08Hz); 9.98(s,1H,N=CH); 10.74 (s,H,NH);13CNMR (100MHz,DMSO-d6):δ 14.71(OCH2CH3); 55.48(OCH3); 64.83(OCH2CH3); 114.00(2C); 116.22(2C); 118.02; 121.17; 126.51; 126.55; 127.66(2C); 128.38(2C); 128.51130.24(2C); 132.54;

141.07; 151.80; 164.01(2C), (arom-C); 146.03 (triazole C3), 150.40 (N=CH), 152.29

triazole C5), 164.26(COO); UV λ_{max} (ε): 256 (32188), 242 (30552), 226 (29240), nm; Anal. Calcd. for C25 H22N4O5 (458.47): C, 65.49; H, 4.84; N, 12.22. Found: C, 65.46; H, 4.92; N, 12.16

2.2. Antimicrobial Activity test and Minimum inhibitory concentration determination (MIC)

Agar well diffusion method a simple susceptiblennes screening test using agar-well diffusion as adapted earlier was used. An agar well diffusion method was used to measure the antimicrobial activities of 6 type (6a-h) compounds, except for 6b and 6i. Bacteria to be tested were seeded on a pre-prepared Muller Hilton agar medium with dilutions of approximately 10 cfu / ml in a Mueller Hinton liquid medium from overnight cultures. Wells of 5 mm in diameter were opened at 2 cm intervals with sterile glass tubing on the streaked media, and (10 µg/50 µL) of each stock solution (containing 250-500 µg / mL of substance) was dropped from each chemical stock solution. Petri dishes containing bacteria were incubated for 24 hours at 35 °C and inhibition zones were measured with a ruler after incubation.

All test microorganisms used are Bacillus subtilis (ATCC11774) (Gram (+) spore bacterium), Yersinia enterocolitca (ATCC27729) enteric bacterium), Bacillus cereus (ATCC11778) (Gram (+) spore bacterium), Staphylococcus aureus (ATCC6538) (Gram (-) enteric bacterium), Esherichia coli (ATCC25922) (Gram (-) enteric bakteri). Pasterulla multicida (ATCC12945) (Gram (-) enteric bacterium, Klebsiella pneumonia (ATCC4352) (Gram (-) encapsulated bacterium), from Microbiological Environmental Protection Laboratories (France). All synthesized compounds were dissolved in dimethylsulfoxide to prepare stock solutions.

3. Result

3.1. Synthesis of compound

We reported nine synthesis of compound totally during our research (Table 1). Our aim is study of antibacterial activity of etoksibenzilidenamino derived 1,2,4-triazoles. We used seven synthesis of compound of them.

Table1: Synthesis, characterization and antimicrobial activity of novel etoksibenzilidenamino derived 1,2,4-triazoles

	R
a	CH_3
b	CH_2CH_3
c	$CH_2CH_2CH_3$
d	$CH_2C_6H_5$
e	$CH_2C_6H_4CH_3$ (*p*-)
f	$CH_2C_6H_4OCH_3$ (*p*-)
g	$CH_2C_6H_4Cl$ (*p*-)
h	$CH_2C_6H_4Cl$ (*m*-)
i	C_6H_5

3.2. Minimum inhibitory concentration determination (MIC)

The zones inhibition diameters and the MIC values indicate that all synthesized compounds 6a-h exhibited an antimicrobial activity against Gram-, Gram+ except for 6b and 6i. Also we applay some antibiotic

(Ampillicin, Neomycin Streptomycin) against the same strain of bacteria. The inhibition zone diameters (IZD) and MIC values are set in figure 2.

The antimicrobial activity of tested compounds was qualitatively and quantitatively assessed by the diskdiffusion. The inhibition zone diameters (IZD) and MIC values are set in Table 2.

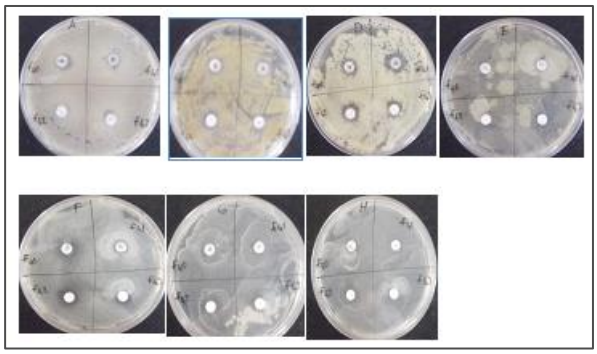

Figure 2: Zone diameters of type Compounds and standards versus bacteria (F40;6a-h)

Table2: Antimicrobial activity of etoksibenzilidenamino derived 1,2,4-triazoles (10 mg/ml)) and The inhibition zone diameters (IZD) and MIC values

Compounds Volume (mm)		Species of Microorganisms						
		Bacillius subtilis A	*Yersinia enterocolitica* C	*Bacillus cereus* D	*Staphlacoccus aureus* E	*Escherichia coli* F	*Pasterulla multicida* G	*Klepsiella pnemonias* H
F40	6a	15	10	11	-	17	18	11
F 41	6c	8	-	15	7	19	21	13
F 42	6d	-	10	13	-	13	17	17
F 43	6e	5	-	10	9	15	13	18
F 44	6f	9	-	11	10	12	16	17
F 45	6g	10	-	12	12	16	15	14
F 46	6h	9	-	11	10	15	-	-
Ampillicin		32	36	35	37	35	36	36
Neomycin		19	16	15	14	17	17	16
Streptomycin		7	13	10	18	15	18	17

4. Conclusion

The synthesis of a series of etoksibenzilidenamino derived 1,2,4-triazoles has been realized with good yields using the CuAAC of a variety of alkynyl heterocycles, alkynyl carbohydrates or alkynes. The structures of the obtained

compounds were confirmed by NMR spectroscopy (1 H and 13C) and mass spectrometry. Overall, the molecules are not showed very antibacterial activity, but our antibiotic showed that important antibacterial activity at in vıtro condition. Many similarity research raported that newly synthesized final compounds were evaluated for their in vitro antibacterial and antifungal activities against four strains each (Eswaran, at all,2009) and then they recorded preliminary results indicated that most of the compounds demonstrated very good antimicrobial activity, comparable to the first line standard drugs. The most effective compounds have exhibited activity at MIC of 6.25 mg/mL.

5. References

Chu D.T., Plattner J.J., Katz L., J. Med. 1996. New directions in antıbacterial research. Chemicial 39-3853-3874.

Chua T., Moore C.L., Perri M.B., Donabedian S.M., Masch W., Vager D., Davis S. L., Lulek K., Zimnicki B., Zervos M.J., J. 2008. Molecular epidemiology of methicillin- resistant Staphylococcus aureus bloodstream isolates in urban Detroit. Journal of Clinical Microbiology, 46- 2345-2352.

Darandale S.N., Mulla N. a., Pansare D.N., Sangshetti J.N., Shinde D.B., Eur. J. Med. 2013. A novel amalgamation of 1,2,3-triazoles, piperidines and thieno pyridine rings and evaluation of their antifungal activity. Chemistry 65 -527-532.

Esvaran S. A. V. Adhikari N., S. Shetty. 2009. Synthesis and antimicrobial activities of novel quinoline derivatives carrying 1,2,4-triazole moiety. European Journal of Medicinal Chemistry 44. 2009. 4637–4647

Fichtali W., Laboudi, E.M. El hadrami, F. El Aroussi, A.Ben-Tama, M.Benlemlıh, S.-E. Stiriba, 2016 Synthesis, characterization and antimicrobial activity of novel benzophenone derived 1,2,3-triazoles. Journal of Materials and Environmental Science, 7 (5) ,2016, 1633-1641.

He Y.-W., Dong C.-Z., Zhao J.-Y., Ma L.-L., Li Y.-H., Aisa H.A., Eur. J. 2014. 1,2,3-Triazole-containing derivatives of rupestonic acid: click-chemical synthesis and antiviral activities against influenza viruses. Journal of Medicinal Chemistry, 76 245-255.

Henry G.E., Jacobs H., Carrington C.M.S., McLean S., Reynolds W.F., 1999. Prenylated Benzophenone Derivatives fromCaribbeanlusiaspecies (Guttiferae). Plukenetiones –G and Xerophenone A.Tetrahedron. 55 1581-1596.

Kamal A., Hussaini S.M.A., Faazil S., Poornachandra Y., Narender Reddy G., Kumar C.G., Rajput V.S., Rani C., Sharma R., Ali Khan I., Babu N.J., 2013.Bioorganic and Medicinal Chemistry Letters, 23 -6842-6846.

Karrer F., Meier H., Pascual A., 2000. Short synthesis of 4-chloro-4′-(chlorodifluoromethoxy)benzophenone Journal of Fluorine Chemistry, 103-81-84.

Kaushik C.P., Lal K., Kumar A., Kumar S., 2014. Synthesis and Antiprotozoal Activity of Cationic 1,4-Diphenyl-1H-1,2,3-triazoles Medicinal Chemistry Research, 23 2995-3004.

Kharb R., Sharma P.C., Yar M.S., J. 2011. Pharmacological significance of triazole scaffold. Journal Enzyme Inhibition and Medicinal Chemistry, 26- 1-21.

Tarik El-Sayed Ali, 2009."Synthesis and antibacterial activity of some new thiadiaza/triazaphospholes, thiadiaza/triaza/tetrazaphosphinines and thiadiaza/tetrazaphosphepines containing 1,2,4-triazinone moiety" European Journal of Medicinal Chemistry S: 44 s. 4539–4546

Thirumurugan P., Matosiuk D., Jozwiak K., 2013. Click chemistry for drug development and diverse chemical-biology applications. Chemical Reviews, 113- 4905-4979.

Ulloora S., Shabaraya R., Adhikari A.V., Bioorg. 2013. Facile synthesis of new imidazo[1,2-a]pyridines carrying 1,2,3-triazoles via click chemistry and their antiepileptic studies. Medicinal Chemistry Letters, 23 -3368-3372.