Journal of Sciences, Islamic Republic of Iran 29(3): 211 - 220 (2018) http://jsciences.ut.ac.ir
University of Tehran, ISSN 1016-1104

In vitro Antibacterial Evaluation of Newly Synthesized Heterocyclic Compounds Against *Streptococcus Pneumoniae*

H. Beyzaei[1] and B. Ghasemi[*2]

[1] *Department of Chemistry, Faculty of Science, University of Zabol, Zabol, Islamic Republic of Iran*
[2] *Young Researchers and Elite Club, Neyshabur Branch, Islamic Azad University, Neyshabur, Islamic Republic of Iran*

Received: 4 November 2016 / Revised: 30 February 2017 / Accepted: 3 June 2017

Abstract

In recent years, the spread of drug-resistant strains of *Streptococcus pneumoniae*, as the most common causes of bacterial respiratory infections, threatens public health. Therefore, the use of new antimicrobial medicines to inhibit this pathogen is an urgent demand. In this research project, the inhibitory effects of thirty recently synthesized compounds including thiazole, thiazolidine, imidazole, tetrahydropyrimidine, oxazolidine and thiazepine derivatives against this bacterium have been studied as well. The broth microdilution method was used to determine the minimum inhibitory concentration (MIC) and the minimum bactericidal concentration (MBC), zone diameters of bacterial growth inhibition were also measured by a disk diffusion method. All the obtained results were compared with antibacterial effects of gentamicin and penicillin antibiotics. Among all investigated compounds, only derivatives 3d, 5a, 5b, 7, 9c, 15, 17c and 17d showed inhibitory effects against *S. pneumonia*. As a result, the most and least effects respectively belonged to thiazole derivative 15 and thiazepine derivative 17c with zone diameters of bacterial growth inhibition= 20.2, 9.3 mm, MIC= 64, 2048 μg/mL and MBC= 128, 4096 μg/mL. Whereas thiazole derivative 15 exhibited a good inhibitory activity against the mentioned pathogen, it can be replaced as a good agent instead of antibiotics.

Keywords: Antibacterial effects; *Streptococcus pneumoniae*; Heterocyclic compounds.

Introduction

Streptococcus pneumoniae is a gram-positive bacterium, the normal flora of the nasopharynx and one of the most common causes of respiratory infections that can be spread to the environment, children and the elderly people are the more prone to infection than the others [1]. In recent years, the spread of antibiotic-resistant strains of this pathogen have been known as a threat for public health all over the world [2]. This global problem has encouraged researchers to identify the new effective antibacterial agents [3, 4].

Various reports of biological properties of heterocyclic compounds particularly thiazole, thiazolidine, imidazole, tetrahydropyrimidine, oxazolidine and thiazepine derivatives have been published by the researchers. Consequently, these derivatives were identified as effective agents against a

[*] Corresponding author: Tel: +985431232186; Fax: +985431232180; Email: behzad.ghasemi99@gmail.com

wide range of pathogenic bacteria, antibacterial effects in comparison to the others properties were furtherly evaluated [5-10].

Among the listed derivatives, the thiazoles have been more considered. In addition to beneficial properties including anticancer, anti-allergic and anti-inflammatory, some compounds were exhibited the powerful inhibitory effects against pathogens such as *Staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa* and *Bacillus subtilis* [11].

Thiazolidine derivatives have been identified to inhibit the colon cancer cell growth, the protozoan parasite, *Trypanosoma*, and pathogenic bacteria and fungi such as *Aspergillus niger*, *P. aeruginosa*, *S. aureus* and *Streptococcus pyogenes* [12, 13].

Inhibitory effects of tetrahydropyrimidines against pathogens such as *Klebsiella pneumoniae*, *P. aeruginosa*, *Aspergillus* and *Candida* have been proven, besides they are effective in the treatment of Alzheimer's disease [14].

Some imidazole derivatives were known as antibacterial agents against *E. coli* and *S. aureus*, they have been also used to inhibit tumor cell growth, *Leishmania* parasite, *Aspergillus* and *Fusarium* fungi [15].

Antibacterial effects of oxazolidine derivatives against *Staphylococcus epidermidis* and *Streptococcus agalactiae* have been exhibited in last researches [9]. They have been used to treat inflammatory diseases and AIDS [16, 17].

Thiazepine derivatives have been introduced as the inhibitors of *Enterobacter aerogenes*, *E. coli* and *Luteus catarrhalis*. Diseases and complications such as hypertension, septic shock and arthritis have been treated by the thiazepines [18].

The essential biological properties such as antibacterial effects of listed derivatives and the extend of antibiotic-resistant strains of *S. pneumonia* have encouraged us to investigate the antimicrobial activities of thirty newly synthesized heterocycles including thiazole, thiazolidine, imidazole, tetrahydropyrimidine, oxazolidine and thiazepine derivatives against standard strains of this pathogenic bacterium.

Materials and Methods

The chemical structure of all synthesized compounds were characterized by single crystal X-ray diffraction (Bruker AXS Smart Apex II CCD diffractometer, Germany), ^{1}H NMR (Bruker AC 100 spectrometer, Germany), ^{13}C NMR (Bruker Avance DRX-500 Fourier transform spectrometer, Germany), IR (Bruker Tensor-27 FT-IR spectrometer, Germany), elemental analysis (Thermo Finnigan Flash EA microanalyzer, USA) and mass spectrometry (Varian Mat CH-7, USA). Solution of all derivatives were prepared in DMSO at initial concentrations of 8192 µg/mL. All tests were repeated three times and the results were expressed as the average of three independent experiments.

General procedure for the synthesis of 2-(imidazolidin or tetrahydropyrimidin-2-ylidene)malononitriles 3a-g

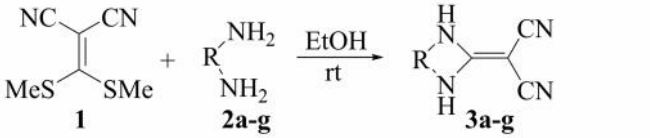
Treatment of 2-[bis(methylthio) methylene] malononitrile (**1**) with diaminoalkanes **2a-g** in ethanol at room temperature for 10-30 min. afforded imidazolidine or tetrahydropyrimidine derivatives **3a-g** (Scheme 1) [19].

*2-(Tetrahydropyrimidin-2(1H)-ylidene)malononitrile (**3a**)*

m.p. 233-234 °C; IR (KBr) *v*: 3275 (NH), 2164 (C≡N), 1620 (C=C) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ: 1.70-1.72 (m, 2H, -CH_2C$\underline{H_2}$$CH_2$-), 3.07-3.11 (m, 4H, -NHC$\underline{H_2}$-), 7.83 (bs, 2H, NH) ppm; ^{13}C NMR (DMSO-d_6) δ: 24.3 (-CH_2C$\underline{H_2}$$CH_2$-), 29.0 (-$\underline{C}(CN)_2$), 51.4 (-NHC$\underline{H_2}$-), 116.7 (C≡N), 155.2 (-$\underline{C}$=C(CN)$_2$) ppm; Anal. Calcd for $C_7H_8N_4$: C, 56.74; H, 5.44; N, 37.82. Found: C, 56.71; H, 5.49, N, 37.80.

*2-(4,4-Dimethylimidazolidin-2-ylidene)malononitrile (**3b**)*

m.p. 328-329 °C; IR (KBr) *v*: 3300 (NH), 2170 (C≡N), 1589 (C=C) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ: 1.77 (s, 6H, CH_3), 3.32 (s, 2H, -NHC$\underline{H_2}$-), 7.70 (bs, 2H, NH) ppm; ^{13}C NMR (DMSO-d_6) δ: 19.6 (CH_3), 28.3 (-$\underline{C}$(CN)$_2$), 50.6 (-$NHCH_2$-), 59.0 (-$\underline{C}$(CH_3)$_2$-), 118.7



R = a: -$CH_2CH_2CH_2$-; b: -$CH_2C(CH_3)_2$-; c: -$CH_2CH(CH_3)$-
d: -CH_2CH_2CH(Et)-; e: -CH_2CH(OH)CH_2-
f: -$CH_2C(CH_3)_2CH_2$; g: 1,2-cyclohexyl

Scheme 1. Synthesis of 2-(imidazolidin or tetrahydropyrimidin-2-ylidene)malononitriles.

(C≡N), 159.4 (-C=C(CN)$_2$) ppm; Anal. Calcd for $C_8H_{10}N_4$: C, 59.24; H, 6.21; N, 34.55. Found: C, 59.18; H, 6.30; N, 34.52.

*2-(4-Methylimidazolidin-2-ylidene)malononitrile (**3c**)*

m.p. 246-247 °C; IR (KBr) *v*: 3260 (NH), 2186 (C≡N), 1601 (C=C) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ: 1.17 (d, 3H, *J* = 6.2 Hz, -CH$_3$), 3.11 (t, 2H, *J* = 9.8 Hz, -NHCH$_2$-), 3.97-3.99 (m, 1H, -CH(CH3)-), 8.16 (bs, 2H, -NH) ppm; ^{13}C NMR (DMSO-d_6) δ: 20.2 (-CH3), 27.5 (-C(CN)$_2$), 50.5 (-NHCH2-), 52.0 (-CH(CH3)-), 117.8 (C≡N), 165.4 (-C=C(CN)$_2$) ppm; Anal. Calcd for C7H8N4: C, 56.74; H, 5.44; N, 37.82. Found: C, 56.66; H, 5.48; N, 37.86.

*2-(4-Ethyltetrahydropyrimidin-2(1H)-ylidene) malononitrile (**3d**)*

m.p. 188-190 °C; IR (KBr) *v*: 3288 (NH), 2172 (C≡N), 1573 (C=C) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ: 0.84 (t, 3H, *J* = 7.4 Hz, -CH$_3$), 1.38-1.40 (m, 2H, -NHCH$_2$CH$_2$-), 1.59-1.64 (m, 2H, -CH$_2$CH$_3$), 3.17-3.20 (m, 2H, -NHCH$_2$-), 3.25-3.27 (m, 1H, -CHEt), 7.58 (bs, 2H, NH) ppm; ^{13}C NMR (DMSO-d_6) δ: 9.4 (-CH$_3$), 23.6 (-CH$_2$CH$_3$), 27.0 (-NHCH$_2$CH$_2$-), 27.7 (-C(CN)$_2$), 36.6 (-NHCH$_2$-), 50.0 (-CHEt), 118.8 (C≡N), 159.2 (-C=C(CN)$_2$) ppm; Anal. Calcd for $C_9H_{12}N_4$: C, 61.34; H, 6.86; N, 31.80. Found: C, 61.38; H, 6.79; N, 31.83.

*2-(5-Hydroxytetrahydropyrimidin-2(1H)-ylidene) malononitrile (**3e**)*

m.p. 278-280 °C; IR (KBr) *v*: 3313 (NH, OH), 2172 (C≡N), 1616 (C=C) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ: 3.06 (d, 4H, *J* = 12.5 Hz, -NHCH$_2$-), 3.97 (m, 1H, -CHOH), 5.28 (bs, 1H, -OH), 7.61 (bs, 2H, -NH) ppm; ^{13}C NMR (DMSO-d_6) δ: 29.2 (-C(CN)$_2$), 44.9 (-NHCH$_2$-), 58.2 (-CHOH), 118.8 (C≡N), 159.0 (-C=C(CN)$_2$) ppm; Anal. Calcd for $C_7H_8N_4O$: C, 51.21; H, 4.91; N, 34.13; O, 9.75. Found: C, 51.18; H, 4.87; N, 34.19; O, 9.76.

*2-(5,5-Dimethyltetrahydropyrimidin-2(1H)-ylidene) malononitrile (**3f**)*

m.p. 308-310 °C; IR (KBr) *v*: 3257 (NH), 2168 (C≡N), 1616 (C=C) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ: 0.92 (s, 6H, -CH$_3$), 2.88 (s, 4H, -CH$_2$), 7.75 (bs, 2H, -NH) ppm; ^{13}C NMR (DMSO-d_6) δ: 23.4 (CH$_3$), 26.4 (-C(CH$_3$)$_2$-), 29.0 (-C(CN)$_2$), 49.7 (CH$_2$), 118.7 (-C≡N), 158.6 (-C=C(CN)$_2$) ppm; Anal. Calcd for $C_9H_{12}N_4$: C, 61.34; H, 6.86; N, 31.80. Found: C, 61.40; H, 6.91; N, 31.69.

*2-(Octahydro-2H-benzo[d]imidazol-2-ylidene) malononitrile (**3g**)*

m.p. 330-331 °C; IR (KBr) *v*: 3253 (NH), 2197 (C≡N), 1600 (C=C) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ: 1.25-1.38 (m, 4H, -CHCH$_2$CH$_2$-), 1.69-1.73 (m, 4H, -CHCH$_2$CH$_2$-), 3.01-3.05 (m, 2H, -NHCH-), 8.44 (bs, 2H, -NH) ppm; ^{13}C NMR (DMSO-d_6) δ: 23.3 (-CHCH$_2$CH$_2$-), 28.4 (-CHCH$_2$CH$_2$-), 63.0 (-NHCH-), 30.0 (-C(CN)$_2$), 117.2 (C≡N), 168.6 (-C=C(CN)$_2$) ppm; Anal. Calcd for $C_{10}H_{12}N_4$: C, 63.81; H, 6.42; N, 29.77. Found: C, 63.78; H, 6.51; N, 29.71.

General procedure for the synthesis of cyclic 1,3-diamines 5a,b

Cyclic 1,3-diamines **5a,b** were synthesized in the reaction of thioamide **4** with 1,3-diaminopropane (**2a**) or 1,2-diaminopropane (**2b**) in ethanol under reflux conditions for 16 and 18 h, respectively (Scheme 2) [20].

*2,2-Bis(1,4,5,6-tetrahydropyrimidin-2-yl)acetonitrile (**5a**)*

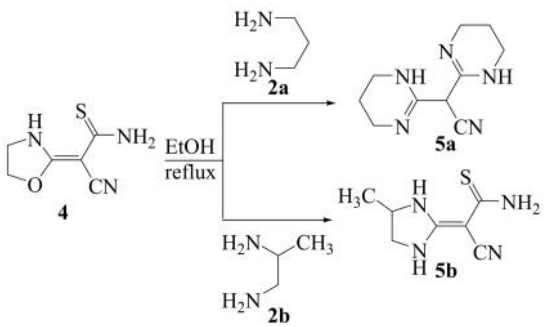
m.p. 209-210 °C; IR (KBr) *v*: 3286 (NH), 2171 (C≡N), 1688 (C=C) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ: 1.76-1.79 (m, 4H, -NHCH$_2$CH$_2$-), 3.16-3.20 (m, 8H, -NHCH$_2$-), 7.70 (bs, 3H, NH, N≡C–CH-) ppm; ^{13}C NMR (DMSO-d_6) δ: 19.6 (-NHCH$_2$CH$_2$-), 29.2 (N≡C–C-), 38.6 (-NHCH$_2$-), 118.7 (C≡N), 159.3 (–N=C–NH–); Anal. Calcd for $C_{10}H_{15}N_5$: C, 58.51; H, 7.37; N, 34.12. Found: C, 58.53; H, 7.27; N, 34.20.

*(Z,E)-2-Cyano-2-(4-methylimidazolidin-2-ylidene) ethanethioamide (**5b**)*

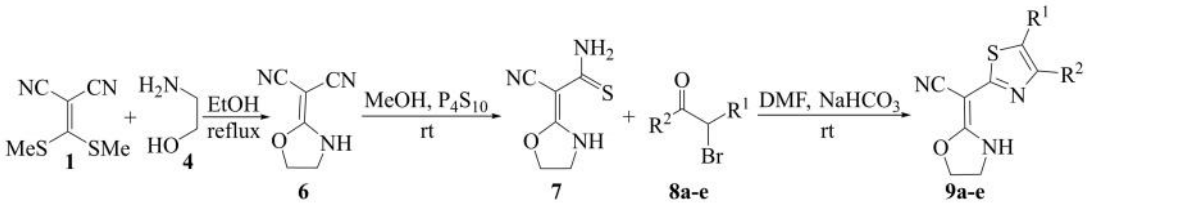
m.p. 189-190 °C; IR (KBr) *v*: 3422 (NH$_2$), 3282 (NH), 2182 (C≡N), 1626 (C=C) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ: 1.21 (d, 3H, *J* = 6.2, -CH$_3$), 3.18 (d, 2H, *J* = 7.5 Hz, -CH$_2$-), 4.04-4.06 (m, 1H, -CHCH$_3$-), 7.29 (bs, 2H, -NH$_2$), 9.13 (bs, 2H, -NH) ppm; ^{13}C NMR (DMSO-d_6) δ: 20.4 (-CH$_3$), 50.2 (-CH$_2$-), 51.2 (-CHCH$_3$), 63.0 (N≡C–C=C-), 118.9 (C≡N), 164.3 (N≡C–C=C-), 188.7 (-C=S) ppm; Anal. Calcd for $C_7H_{10}N_4S$: C, 46.13; H, 5.53; N, 30.74. Found: C, 46.08; H, 5.61; N, 30.81.

General procedure for the synthesis of (oxazolidin-2-ylidene)thiazoles 9a-e

The mixture of compound **1** and ethanolamine (**4**) in ethanol was heated for 5 h to give oxazolidine **6**. Then, this product was thionated in the reaction with P_4S_{10} in absolute methanol for 2 h at room temperature to afford



Scheme 2. Synthesis of cyclic 1,3-diamines.



a: R^1 = H, R^2 = CH_3; b: R^1 = $COCH_3$, R^2 = CH_3; c: R^1 = CO_2Et, R^2 = CH_3; d: R^1 = H, R^2 = 4-Cl-C_6H_4; e: R^1 = H, R^2 = 4-Br-C_6H_4

Scheme 3. Synthesis of (oxazolidin-2-ylidene)thiazoles.

thoamide **7**. Finally, thiazoles **9a-e** were prepared in the reaction of compound **7** with *α*-bromocarbonyls **8a-e** in DMF at room temperature for 2 h (Scheme 3) [21].

(E)-2-Cyano-2-(oxazolidin-2-ylidene)ethanethioamide (7)

m.p. 290-291 °C; IR (KBr) *v*: 3455, 3170 (NH, NH_2), 2197 (C≡N), 1622 (C=C) cm^{-1}; ^{1}H NMR (Aceton-d_6) δ: .05 (t, *J* = 8.4 Hz, 2H, NHC$\underline{H}_2$), 4.77 (t, *J* = 8.4 Hz, 2H, OCH_2), 7.48 (bs, 2H, NH_2), 11.38 (bs, 1H, NH) ppm; MS (EI, m/z): 169 (M^+, 6), 148 (35), 107 (29), 74 (100), 53 (71); Anal. Calcd for $C_6H_7N_3OS$: C, 42.59; H, 4.17; N, 24.83. Found: C, 42.66; H, 4.25; N, 24.80.

*(E)-2-(4-Methylthiazol-2-yl)-2-(oxazolidin-2-ylidene) acetonitrile (**9a**)*

m.p. 164-165 °C; IR (KBr) *v*: 3426 (NH), 2202 (C≡N), 1613 (C=C) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ: 2.30 (s, 3H, CH_3), 3.83 (t, *J* = 8.5 Hz, 2H, NHC$\underline{H}_2$), 4.66 (t, *J* = 8.5 Hz, 2H, OCH_2), 6.83 (s, 1H, C=C-H), 9.48 (bs, 1H, NH) ppm; MS (EI, m/z): 207 (M^+, 13), 205 (100), 145 (36), 133 (16), 67 (28); Anal. Calcd for $C_9H_9N_3OS$: C, 52.16; H, 4.38; N, 20.27. Found: C, 52.23; H, 4.45; N, 20.24.

*(E)-2-(5-Aacetyl-4-methylthiazol-2-yl)-2-(oxazolidin-2-ylidene)acetonitrile (**9b**)*

m.p. 296-297 °C; IR (KBr) *v*: 3441 (NH), 2200 (C≡N), 1648 (C=O), 1606 (C=C) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ: 2.44 (s, 3H, $COCH_3$), 2.61 (s, 3H, CH_3), 3.88 (t, *J* = 8.5 Hz, 2H, NHC$\underline{H}_2$), 4.71 (t, *J* = 8.5 Hz, 2H, OCH_2), 9.75 (bs, 1H, NH) ppm; MS (EI, m/z): 249 (M^+, 7), 233 (42), 173 (35), 93 (15), 46 (100); Anal. Calcd for $C_{11}H_{11}N_3O_2S$: C, 53.00; H, 4.45; N, 16.86. Found: C, 53.08; H, 4.49; N, 16.80.

*Ethyl 2-((E)-cyano(oxazolidin-2-ylidene)methyl) -4-methylthiazole-5 carboxylate (**9c**)*

m.p. 247-248 °C; IR (KBr) *v*: 3447 (NH), 2204 (C≡N), 1696 (C=O), 1612 (C=C) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ: 1.23 (t, *J* = 7.0 Hz, 3H, $CH_2C\underline{H}_3$), 2.55 (s, 3H, CH_3), 3.57 (t, *J* = 8.5 Hz, 2H, NHC$\underline{H}_2$), 4.18 (q, *J* = 7.0 Hz, 2H, $OC\underline{H}_2CH_3$), 4.68 (t, *J* = 8.5 Hz, 2H, OCH_2), 9.67 (bs, 1H, NH) ppm; MS (EI, m/z): 279 (M^+, 22), 233 (38), 135 (74), 93 (24), 55 (100); Anal. Calcd for $C_{12}H_{13}N_3O_3S$: C, 51.60; H, 4.69; N, 15.04. Found: C, 51.59; H, 4.75; N, 14.97.

*((E)-2-(4-(4-Chlorophenyl)thiazol-2-yl)-2-(oxazolidin -2-ylidene)acetonitrile (**9d**)*

m.p. 240-241 °C; IR (KBr) *v*: 3423 (NH), 2203 (C≡N), 1617 (C=C) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ: 3.90 (t, *J* = 8.4 Hz, 2H, NHC$\underline{H}_2$), 4.68 (t, *J* = 8.4 Hz, 2H, OCH_2), 7.44 (d, *J* = 8.3 Hz, 2H, H-3,5 Ph), 7.76 (s, 1H, C=C-H), 8.08 (d, *J* = 8.3 Hz, 2H, H-2,6 Ph), 9.26 (bs, 1H, NH) ppm; MS (EI, m/z): 304 (M^+, 18), 246 (69), 112 (48), 68 (100), 39 (71); Anal. Calcd for $C_{14}H_{10}ClN_3OS$: C, 55.35; H, 3.32; N, 13.83. Found: C, 55.29; H, 3.40; N, 13.88.

*((E)-2-(4-(4-Bromophenyl)thiazol-2-yl)-2-(oxazolidin -2-ylidene)acetonitrile (**9e**)*

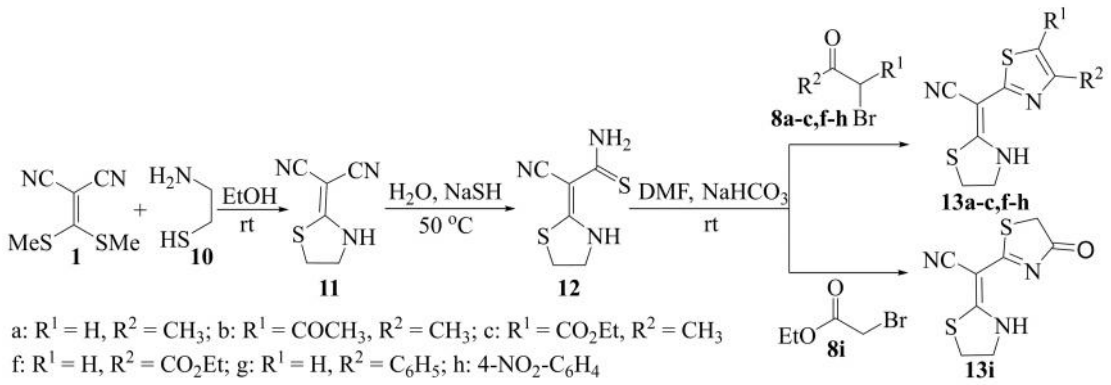
m.p. 245-246 °C; IR (KBr) *v*: 3433 (NH), 2203 (C≡N), 1610 (C=C) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ: 3.89 (t, *J* = 8.3 Hz, 2H, NHC$\underline{H}_2$), 4.68 (t, *J* = 8.3 Hz, 2H, OCH_2), 7.56 (d, *J* = 8.3 Hz, 2H, H-2,6 Ph), 7.76 (s, 1H, C=C-H), 8.00 (d, *J* = 8.3 Hz, 2H, H-3,5 Ph), 9.25 (bs, 1H, NH) ppm; MS (EI, m/z): 348 (M^+, 15), 302 (44), 288 (31), 229 (100), 181 (84), 124 (58); Anal. Calcd for $C_{14}H_{10}BrN_3OS$: C, 48.29; H, 2.89; N, 12.07. Found: C, 48.26; H, 2.93; N, 12.02.

General procedure for the synthesis of (thiazolidin-2-ylidene)thiazoles 13a-c,f-i

A suspension of compound **1** and cysteamine (**10**) in ethanol was stirred at room temperature for 5 h to give thiazolidine **11**. Then, thionation of this product was occurred in the reaction with NaSH in water for 22 h at 50 ^oC to obtain thoamide **12**. Finally, thiazoles **9a-d** were prepared in the reaction of compound **12** with *α*-bromocarbonyls **8a-c,f-i** in DMF at room temperature for 2-8 h (Scheme 4) [22].

*2-(Thiazolidin-2-ylidene)malononitrile (**11**)*

m.p. 209-212 °C; IR (KBr) *v*: 3195 (NH), 2207 (C≡N), 1571 (C=C) cm^{-1}; ^{1}H NMR ($CDCl_3$) δ: 3.48 (t, *J* = 6.9 Hz, 2H, SCH_2), 3.99 (t, *J* = 6.9 Hz, 2H, NHC$\underline{H}_2$), 6.84 (s, 1H, NH) ppm; MS (EI, m/z): 151 (M^+, 14), 124 (72), 119 (100), 88 (31), 63 (64); Anal. Calcd for $C_6H_5N_3S$: C, 47.66; H, 3.34; N, 27.79. Found: C, 47.56; H, 3.43; N, 27.71.



Scheme 4. Synthesis of (thiazolidin-2-ylidene)thiazoles.

*(E)-2-Cyano-2-(thiazolidin-2-ylidene)ethanethioamide (**12**)*

m.p. 217-219 °C; IR (KBr) *v*: 3435, 3320 (NH, NH_2), 2182 (C≡N), 1570 (C=C) cm^{-1}; ^{1}H NMR (acetone-d_6) δ: 3.61 (t, *J* = 7.1 Hz, 2H, SCH_2), 4.32 (t, *J* = 7.1 Hz, 2H, $NHCH_2$), 7.50 (s, 2H, NH_2), 12.17 (1H, s, NH) ppm; ^{13}C NMR (DMSO-d_6) δ: 30.2 (SCH_2), 52.8 ($NHCH_2$), 75.4 (-NH-C=C-), 119.4 (C≡N), 175.5 (-NH-C=C-), 189.5 (C=S) ppm; MS (EI, m/z): 185 (M^+, 3), 153 (72), 107 (555), 90 (78), 59 (100); Anal. Calcd for $C_6H_7N_3S_2$: C, 38.90; H, 3.81; N, 22.68. Found: C, 38.95; H, 3.74; N, 22.76.

*(E)-2-(4-Methylthiazol-2-yl)-2-(thiazolidin-2-ylidene) acetonitrile (**13a**)*

m.p. 166-168 °C; IR (KBr) *v*: 3438 (NH), 2180 (C≡N), 1563 (C=C) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ: 2.32 (s, 3H, CH_3), 3.47 (t, *J* = 7.7 Hz, 2H, SCH2), 4.01 (t, *J* = 7.7 Hz, 2H, NHCH2), 6.91 (s, 1H, C=C-H), 9.94 (s, 1H, NH) ppm; MS (EI, m/z): 223 (M^+, 8), 162 (70), 149 (5), 71 (24), 28 (100); Anal. Calcd for C9H9N3S2: C, 48.41; H, 4.06; N, 18.82. Found: C, 48.40; H, 4.10; N, 18.74.

*(E)-2-(5-Acetyl-4-methylthiazol-2-yl)-2-(thiazolidin-2-ylidene)acetonitrile (**13b**)*

m.p. 255-257 °C; IR (KBr) *v*: 3421 (NH), 2191 (C≡N), 1648 (C=O), 1557 (C=C) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ: 2.52 (s, 3H, $COCH_3$), 2.65 (s, 3H, CH_3), 3.52 (t, *J* = 7.7 Hz, 2H, SCH_2), 4.12 (t, *J* = 7.7 Hz, 2H, $NHCH_2$), 10.13 (s, 1H, NH) ppm; MS (EI, m/z) 265 (M^+, 26), 204 (46), 162 (20), 61 (69), 43 (100); Anal. Calcd for $C_{11}H_{11}N_3OS_2$: C, 49.79; H, 4.18; N, 15.84. Found: C, 49.68; H, 4.02; N, 15.60.

*Ethyl 2-((E)-cyano(thiazolidin-2-ylidene)methyl)-4-methylthiazole-5 carboxylate (**13c**)*

m.p. 253-255 °C; IR (KBr) *v*: 3443 (NH), 2,190 (C≡N), 1706 (C=O), 1562 (C=C) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ: 1.26 (t, *J* = 7.1 Hz, 3H, CH_2CH_3), 2.60 (s, 3H, CH_3), 3.57 (t, *J* = 7.6 Hz, 2H, SCH_2), 4.06 (t, *J* = 7.6 Hz, 2H, $NHCH_2$), 4.16 (q, *J* = 7.1 Hz, 2H, OCH_2), 10.13 (s, 1H, NH) ppm; MS (EI, m/z): 295 (M^+, 9), 234 (18), 151 (13), 71 (25), 29 (100); Anal. Calcd for $C_{12}H_{13}N_3O_2S_2$: C, 48.80; H, 4.44; N, 14.23. Found: C, 48.76; H, 4.27; N, 14.05.

*Ethyl2-[(E)-cyano(thiazolidin-2-ylidene)methyl] thiazole-4-carboxylate (**13f**)*

m.p. 264-267 °C; IR (KBr) *v*: 3430 (NH), 2186 (C≡N), 1597 (C=O), 1561 (C=C) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ: 1.34 (t, *J* = 6.6 Hz, 3H, CH_3), 3.52 (t, *J* = 7.2 Hz, 2H, SCH_2), 4.12 (t, *J* = 7.2 Hz, 2H, $NHCH_2$), 4.31 (q, *J* = 6.6 Hz, 2H, OCH_2), 8.24 (s, 1H, C=C-H), 9.93 (s, 1H, NH); MS (EI, m/z): 281 (M^+, 11), 278 (100), 233 (78), 173 (81), 146 (34), 61 (73); Anal. Calcd for $C_{11}H_{11}N_3O_2S_2$: C, 46.96; H, 3.94; N, 14.94. Found: C, 47.07; H, 4.02; N, 14.90. The X-ray diffraction data and details for crystals of thiazole **13f** can be obtained free of charge from the Cambridge Crystallographic Data Center *via* www.ccdc.cam.ac.uk/conts/retrieving.html.

*(E)-2-(4-Phenylthiazol-2-yl)-2-(thiazolidin-2-ylidene) acetonitrile (**13g**)*

m.p. 181-184 °C; IR (KBr) *v*: 3438 (NH), 2187 (C≡N), 1563 (C=C) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ: 3.53 (t, *J* = 7.9 Hz, 2H, SCH_2), 4.12 (t, *J* = 7.9 Hz, 2H, $NHCH_2$), 8.05 (m, 5H, Ph), 7.85 (s, 1H, C=C-H), 9.81 (s, 1H, NH) ppm; MS (EI, m/z): 285 (M^+, 24), 283 (100), 224 (98), 134 (97), 61 (87); Anal. Calcd for $C_{14}H_{11}N_3S_2$: C, 58.92; H, 3.88; N, 14.73. Found: C, 58.94; H, 3.77; N, 14.41.

*(E)-2-(4-(4-Nitrophenyl)thiazol-2-yl)-2-(thiazolidin-2-ylidene)acetonitrile (**13h**)*

m.p. 264-267 °C; IR (KBr) *v*: 3433 (NH), 2193(C≡N), 1569 (C=C) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ: 3.62 (t, *J* = 7.8 Hz, 2H, SCH_2), 4.13 (t, *J* = 7.8 Hz, 2H, $NHCH_2$), 8.15 (s, 1H, C=C-H), 8.30 (d, 2H, *J* = 7.0 Hz, H-2,6 Ph), 8.34 (d, 2H, *J* = 7.0 Hz, H-3,5 Ph), 9.65 (s, 1H, NH) ppm; MS (EI, m/z): 330 (M^+, 15), 328 (100),

267 (75), 195 (27), 133 (52), 61 (83); Anal. Calcd for $C_{14}H_{10}N_4O_2S_2$: C, 50.90; H, 3.05; N, 16.96. Found: C, 51.12; H, 2.97; N, 17.02.

*(2E)-2-(4,5-Dihydro-4-oxothiazol-2-yl)-2-(thiazolidin -2-ylidene)acetonitrile (**13i**)*

m.p. 248-252 °C; IR (KBr) *v*: 3422 (NH), 2193 (C≡N), 1688 (C=O), 1573 (C=C) cm^{-1}; ^1H NMR (DMSO-d_6) δ: 3.63 (t, J = 7.8 Hz, 2H, SCH_2), 4.01 (s, 2H, $COCH_2$), 4.12 (t, J = 7.8 Hz, 2H, $NHC\underline{H_2}$), 10.53 (s, 1H, NH) ppm; MS (EI, m/z): 225 (M^+, 21), 176 (37), 150 (94), 109 (28), 46 (100); Anal. Calcd for $C_8H_7N_3OS_2$: C, 42.65; H, 3.13; N, 18.65. Found: C, 42.74; H, 3.00; N, 18.46.

synthesis of thioamide 15

The mixture of benzo[*d*]thiazole **14** and P_4S_{10} in absolute ethanol was heated under reflux for 5 h to synthesize thioamide **15** (Scheme 5) [23].

*(E)-2-(Benzo[d]thiazol-2(3H)-ylidene)-2-cyano-ethanethioamide (**15**)*

m.p. 251-252 °C; IR (KBr) *v*: 3431, 3295 (NH, NH_2), 2173 (C≡N), 1615 (C=S) cm^{-1}; ^1H NMR (DMSO-d_6) δ: 7.54-7.60 (m, 3H, H-1,2,3 Ph), 7.85 (d, J = 7.2 Hz, 1H, H-4 Ph), 8.49 (bs, 2H, NH_2), 12.97 (bs, 1H, NH) ppm; MS (EI, m/z): 233 (M^+, 11), 202 (63), 171 (33), 129 (100), 92 (71); Anal. Calcd for $C_{10}H_7N_3S_2$: C, 51.48; H, 3.02; N, 18.01. Found: C, 51.40; H, 2.99; N, 18.06.

General Procedure for the Synthesis of thiazepines 17a-d

A solution of 2-((arylamino)methylene) malononitriles **16a-d** and cysteamine (**10**) in DMF as solvent was heated at 60 °C for 12 h to obtain tetrahydro-1,4-thiazepines **17a-d** (Scheme 6) [24].

*(5Z,7Z)-7-(4-Chlorophenylimino)-5-amino-2,3,4,7-tetrahydro-1,4-thiazepine-6-carbonitrile (**17a**)*

m.p. 251-252 °C; IR (KBr) *v*: 3300, 3231 (NH, NH_2), 2197 (C≡N), 1602 (C=N) cm^{-1}; ^1H NMR (Acetone-d_6) δ: 3.00 (t, J = 6.6 Hz, 2H, SCH_2), 3.72 (t, J = 6.6 Hz, 2H, $NHC\underline{H_2}$), 7.04 (s, 2H, NH_2), 7.19 (d, 2H, J = 7.0 Hz, H-2,6 Ph), 7.34 (d, 2H, J = 7.0 Hz, H-3,5 Ph), 8.71 (s, 1H, NH) ppm; MS (EI, m/z): 278 (M^+, 8), 164 (41), 147 (77), 111 (12), 56 (100); Anal. Calcd for $C_{12}H_{11}ClN_4S$: C, 51.70; H, 3.98; N, 20.10. Found: C, 51.56; H, 3.89; N, 20.21.

*(5Z,7Z)-7-(4-Bromophenylimino)-5-amino-2,3,4,7-tetrahydro-1,4-thiazepine-6-carbonitrile (**17b**)*

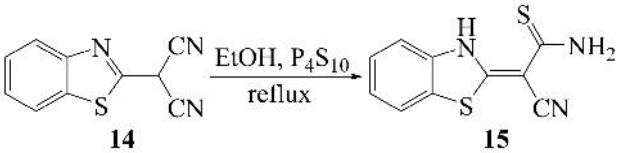
m.p. 264-265 °C; IR (KBr) *v*: 3270, 3247 (NH, NH_2), 2189 (C≡N), 1595 (C=N) cm^{-1}; ^1H NMR (Acetone-d_6) δ: 3.00 (t, J = 6.7 Hz, 2H, SCH_2), 3.72 (t, J = 6.7 Hz, 2H, $NHC\underline{H_2}$), 7.02 (s, 2H, NH_2), 7.22 (d, 2H, J = 7.3 Hz, H-2,6 Ph), 7.54 (d, 2H, J = 7.3 Hz, H-3,5 Ph), 8.74 (s, 1H, NH) ppm; MS (EI, m/z): 323 (M^+, 5), 266 (71), 192 (68), 132 (67), 88 (70), 42 (100); Anal. Calcd for $C_{12}H_{11}BrN_4S$: C, 44.59; H, 3.43; N, 17.33. Found: C, 44.64; H, 3.47; N, 17.47.

*(5Z,7Z)-7-(4-Nitrophenylimino)-5-amino-2,3,4,7-tetrahydro-1,4-thiazepine-6-carbonitrile (**17c**)*

Decomp. 243-244 °C; IR (KBr) *v*: 3324, 3215 (NH, NH_2), 2208 (C≡N), 1648 (C=N) cm^{-1}; ^1H NMR (Acetone-d_6) δ: 3.02 (t, J = 6.4 Hz, 2H, SCH_2), 3.78 (t, J = 6.4 Hz, 2H, $NHC\underline{H_2}$), 7.24 (d, 2H, J = 8.5 Hz, H-2,6 Ph), 7.39 (s, 2H, NH_2), 7.45 (d, 2H, J = 8.5 Hz, H-3,5 Ph), 9.13 (s, 1H, NH) ppm; MS (EI, m/z): 289 (M^+, 5), 253 (98), 177 (72), 107 (70), 60 (100); Anal. Calcd for $C_{12}H_{11}N_5O_2S$: C, 49.82; H, 3.83; N, 24.21. Found: C, 49.94; H, 3.75; N, 24.18.

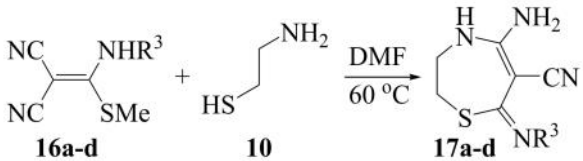
*(5Z,7Z)-5-Amino-2,3,4,7-tetrahydro-7-(phenylimino) -1,4-thiazepine-6-carbonitrile (**17d**)*

m.p. 127-128 °C; IR (KBr) *v*: 3312, 3279 (NH, NH_2), 2192 (C≡N), 1579 (C=N) cm^{-1}; ^1H NMR (Acetone-d_6) δ: 2.93 (t, J = 6.8 Hz, 2H, SCH_2), 3.64 (t, J = 6.8 Hz, 2H, $NHC\underline{H_2}$), 6.90 (s, 2H, NH_2), 7.21-7.29 (m, 5H, Ph), 8.62 (s, 1H, NH) ppm; MS (EI, m/z): 244 (M^+, 9), 214



14 **15**

Scheme 5. Synthesis of 2-cyanoethanethioamide.



16a-d **10** **17a-d**

R^3 = a: 4-Cl-C_6H_4; b: 4-Br-C_6H_4; c: 4-NO_2-C_6H_4; d: C_6H_5

Scheme 6. Synthesis of tetrahydro-1,4-thiazepines.

(62), 183 (100), 153 (83), 114 (59), 78 (91); Anal. Calcd for $C_{12}H_{12}N_4S$: C, 58.99; H, 4.95; N, 22.93. Found: C, 59.12; H, 4.88; N, 22.79.

Preparation of the bacterial suspension

S. pneumoniae (PTCC 1240) was prepared from the Persian Type Culture Collection (PTCC), Tehran, Iran. Bacterium was cultured on blood agar (Merck, Germany), and then incubated for 24 h at 37 °C in the atmosphere of 5% CO_2. Finally, under sterile conditions, a bacterial suspension at concentration of 0.5 McFarland (1.5×10^8 CFU/mL) in Brain-Heart Infusion Broth (Merck, Germany) was obtained using Jenway 6405 UV-Visible spectrophotometer (United Kingdom), which used as a storage source.

Determination of the minimum inhibitory concentration (MIC)

The MIC tests are carried out according to CLSI broth microdilution method in 96-well microliter plates. 90 µL of BHI broth medium was added to all the wells in each row. Then, 100 µL of solution of each derivative in DMSO with concentration of 8192, 4096, 2048, 1024, 512, 256, 128, 64, 32, 16 and 8 µg/mL was added to them, respectively (for antibiotics, concentrations of 256, 128, 64, 32, 16, 8, 4, 2, 1, 0/5 and 0/25 µg/mL were used). 100 µL of DMSO as a negative control was added to the twelve well. Finally, 10 µL of bacterial suspension was added to each well. The plates were incubated with shaking at 100 rpm for 24 h at 37 °C in an atmosphere of 5% CO_2. The MIC was determined as the lowest concentration of derivatives and antibiotics at which no visible bacterial growth observed [25].

Determination of the minimum bactericidal concentration (MBC)

Samples of all wells that showed no growth in the MIC test were cultured in blood agar medium (Merck, Germany), then, were incubated for 24 h at 37 °C under 5% CO_2. The lowest concentration of derivatives or antibiotics that bacteria could not grow was reported as MBC [25].

Measurement of zone diameters of bacterial growth inhibition

10 µL of a bacterial suspension of storage source was spread on the blood agar plates using a sterile swab. Then, sterile blank discs were placed on this culture medium at appropriate intervals. 10 µL of initial concentrations for derivatives and antibiotics (10 µL of DMSO as negative control) were poured onto these disks, zone diameters of bacterial growth inhibition were measured by caliper after incubation for 24 h at 37 °C under 5% CO_2. The results of growth inhibition zone diameter are presented as ± meaning the standard deviation, SPSS software version 22 was used for data analysis [25].

Results and Discussion

Among thirty synthesized compounds in this study, only imidazole **5b**, tetrahydropyrimidines **3d** and **5a**, oxazolidines **9c** and **7**, thiazole **15**, thiazepines **17c** and **17d** had an inhibitory effect on *S. pneumonia*. According to the results shown in Table 1, the order of inhibitory power was found to be **15**>**7**>**17d**>**3d**>**5b**>**9c**>**5a**>**17c**. The highest inhibitory effect was observed for thiazole **15** with zone diameter of inhibition = 20.2 mm, MIC = 64 µg/mL and MBC = 128 µg/mL. Thiazepine derivative **17c** had the lowest conclusion with zone diameter of inhibition, MIC and MBC values of 9.3 mm, 2048 µg/mL and 4096 µg/mL, respectively. In comparison to other compounds, penicillin was shown the best result in antibiogram test with MIC = 0.5 and 1 µg/mL and MBC = 1 µg/mL.

In this study, only some evaluated derivatives had inhibitory effects on *S. pneumonia* which showed difference in antibacterial properties of derivatives in a classification, these differences have been also found in recent researches [5-10].

The antibacterial effects of thiazole derivative **15**, with the highest inhibitory activity in comparison with other derivatives, on *S. aureus* and *E. coli* have been proved in our previous studies [26]. It has been found that thiazole derivatives can prevent bacterial growth through inhibition of enzymatic activities of DNA gyrase (it catalyzes the ATP-dependent negative super-coiling of double-stranded closed-circular DNA) or KAS III (it participates in fatty acid biosynthesis) [10, 27]. In the same research, the inhibitory effects of thiazole derivatives were examined on *S. pneumoniae* [28].

The inhibitory effects of oxazolidine **7**, which showed the highest activity on this bacterium after thiazole **15**, has probably exacerbated due to the presence of the thioamide functional group [29]. Xue and colleagues evaluated the effects of some derivatives oxazolidine on penicillin-resistant *S. pneumoniae* as well [30].

In this study, the inhibitory effects of imidazole derivative **5b** were also observed on *S. pneumonia*. The inhibitory power of some imidazoles against *S. aureus*, *Salmonella typhi*, *E. coli* and *Pseudomonas* have been shown by the measuring zone diameters of growth inhibition [31, 32]. Some researchers believe that antibacterial effects of these derivatives are related to

Table 1. Antibacterial effects of derivatives and antibiotics against *Streptococcus pneumoniae*

Derivatives / Antibiotics	Zone diameter of inhibition (mm)	MIC (µg/mL)	MBC (µg/mL)
3a	-	-	-
3b	-	-	-
3c	-	-	-
3d	14.0±4.1	1024	2048
3e	-	-	-
3f	-	-	-
3g	-	-	-
5a	10.0±1.2	2048	4096
5b	13.0±2.1	512	1024
6	-	-	-
7	16.0±1.3	256	512
9a	-	-	-
9b	-	-	-
9c	11.0±5.1	2048	4096
9d	-	-	-
9e	-	-	-
11	-	-	-
12	-	-	-
13a	-	-	-
13b	-	-	-
13c	-	-	-
13f	-	-	-
13g	-	-	-
13h	-	-	-
13i	-	-	-
15	20.0±2.2	64	128
17a	-	-	-
17b	-	-	-
17c	9.0±3.3	2048	4096
17d	12.0±4.1	2048	2048
Penicillin	26.0±3.1	0/5	1
Gentamycin	22.0±4.2	4	8

—: indicates no inhibitory effect at maximum concentration

inhibition of lipid synthesis or the enzyme dihydrofolate reductase (DHFR) [33].

Tetrahydropyrimidine derivatives have been introduced as calcium channel blockers or CD4 glycoprotein inhibitors which were used in the treatment of cardiovascular and neurological diseases and AIDS [34]. But, the antibacterial effects of these derivatives have recently been considered, hence, their possible mechanism of impact on bacteria has not been identified yet. The antibacterial effects of some tetrahydropyrimidine derivatives were evaluated against *S. pyogenes*, *S. aureus* and *B. subtilis* [35].

Antibacterial activities of thiazepine derivatives have been proven against pathogens, but up to now, researchers have not speculated a theory for their possible mechanisms of action [36].

Conclusion

In this study, antibacterial effects of some newly synthesized imidazole, tetrahydropyrimidine, oxazolidine, thiazole and thiazepine derivatives were proved against standard strains of *S. pneumonia*. Therefore, study of their antibacterial activities against drug-resistant strains of *S. Pneumoniae* and toxicity effects of synthesized derivatives on laboratory animals are suggested for the next researches.

Acknowledgments

The authors would like to thank members of the University of Zabol especially Dr Mohammad Allahbakhsh, deputy of research and technology, and Dr Mansour Ghaffari-Moghaddam, dean of the faculty of science, for their support and assistance in various stages of the project.

References

1. Zielnik-Jurkiewicz B. and Bielicka A. Antibiotic resistance of Streptococcus pneumoniae in children with

acute otitis media treatment failure. *Int. J. Pediatr. Otorhinolaryngol*. **79** (12): 2129-2133 (2015).
2. Nakano S., Fujisawa T., Ito Y., Chang B., Suga S. and Noguchi T. Serotypes, antimicrobial susceptibility, and molecular epidemiology of invasive and non-invasive Streptococcus pneumoniae isolates in paediatric patients after the introduction of 13-valent conjugate vaccine in a nationwide surveillance study conducted in Japan in 2012-2014. *Vaccine* **34** (1): 67-76 (2016).
3. Yang X. Y., Zhang L., Liu J., Li N., Yu G. and Cao K. Proteomic analysis on the antibacterial activity of a Ru(II) complex against Streptococcus pneumoniae. *J. Proteomics* **115**: 107-116 (2015).
4. Shariatifar N., Rezae M., Sayadi M., Mosha M. H., Saeedi M. and Mohammadhosseini N. In-vitro antibacterial evaluation of some fluoroquinolone derivatives against food borne bacteria. *J. Sci. I. R. Iran* **27** (2): 129-133 (2016).
5. Khalil A. M., Berghot M. A. and Gouda M. A. Synthesis and antibacterial activity of some new thiazole and thiophene derivatives. *Eur. J. Med. Chem.* **44** (11): 4434-440 (2009).
6. Desai N., Dodiya A. and Shihory N. Synthesis and antimicrobial activity of novel quinazolinone–thiazolidine–quinoline compounds. *J. Saudi Chem. Soc*. **17** (3): 259-267 (2013).
7. Cheng Y., Yin M., Ren X., Feng Q., Wang J. and Zhou Y. A coordination polymeric gelator basedon Ag (I) and 2, 7-bis (1-imidazole) fluorene: Synthesis, characterization, gelation and antibacterial properties. *Mater. Lett*. **139**: 141-144 (2015).
8. Aridoss G., Amirthaganesan S. and Jeong Y. T. Synthesis, crystal and antibacterial studies of diversely functionalized tetrahydropyridin-4-ol. *Bioorg. Med. Chem. Lett*. **20** (7): 2242-2249 (2010).
9. Ammazzalorso A., Amoroso R., Bettoni G., Fantacuzzi M., De-Filippis B. and Giampietro L. Synthesis and antibacterial evaluation of oxazolidin-2-ones structurally related to linezolid. *Farmaco* **59** (9): 685-690 (2004).
10. Malik G. M., Tailor J. H., Zadafiya S. K. and Rajani D. Synthesis and biological activity of triazolo derivative of Dibenzothiazepine. *Chem. Biol. Interface* **5** (3): 208-218 (2015).
11. Cheng K., Xue J. Y. and Zhu H. L. Design, synthesis and antibacterial activity studies of thiazole derivatives as potent ecKAS III inhibitors. *Bioorg. Med. Chem. Lett*. **23** (14): 4235-4238 (2013).
12. Moreira T. L., Barbosa A. F., Veiga-Santos P., Henriques C., Henriques-Pons A. and Galdino S.L. Effect of thiazolidine LPSF SF29 on the growth and morphology of Trypanosoma cruzi. *Int. J. Antimicrob. Agents* **41** (2): 183-187 (2013).
13. Desai N. C. and Dodiya A. M. Synthesis, characterization and antimicrobial screening of quinoline based quinazolinone-4-thiazolidinone heterocycles. *Arab. J. Chem*. **7** (6): 906-913 (2014).
14. Ghasemi B., Beyzaei H. and Majidiani H. A comparative study on the antibacterial effects of some newly synthesized thiazole, imidazolidine and tetrahydropyrimidine derivatives against Bacillus cereus and Salmonella typhimurium. *Pharm. Sci*. **22** (1): 54-59 (2016).
15. Ghasemi B., Beyzaei H. and Hashemi H. Study of antibacterial effect of novel thiazole, imidazole, and tetrahydropyrimidine derivatives against Listeria monocytogenes. *Ann. Mil. Health Sci. Res.* **13** (3): 101-105 (2015).
16. Park S. J., Kang S. H., Kang Y. K., Eom Y. B., Koh K. O. and Kim D. Y. Inhibition of homodimerization of toll-like receptor 4 by 4-oxo-4-(2-oxo-oxazolidin-3-yl)-but-2-enoic acid ethyl ester. *Int. Immunopharmacol*. **11** (1): 19-22 (2011).
17. Deng B. L., Zhao Y., Hartman T. L., Watson K., Buckheit R.W. and Pannecouque C. Synthesis of alkenyldiarylmethanes (ADAMs) containing benzo[*d*]isoxazole and oxazolidin-2-one rings, a new series of potent non-nucleoside HIV-1 reverse transcriptase inhibitors. *Eur. J. Med. Chem*. **44** (3): 1210-1214 (2009).
18. Ansari F. L., Iftikhar F., Ul-Haq I., Mirza B., Baseer M. and Rashid U. Solid-phase synthesis and biological evaluation of a parallel library of 2,3-dihydro-1,5-benzothiazepines. *Bioorg. Med. Chem*. **16** (16): 7691-7697 (2008).
19. Beyzaei H., Aryan R. and Gomroki M. Synthesis of novel heterocyclic 2-(2-ylidene)malononitrile derivatives. *Org. Chem. Indian J*. **11** (1): 3-10 (2015).
20. Beyzaei H., Aryan R. and Keshtegar Z. Synthesis of new imidazolidine and tetrahydropyrimidine derivatives. *Adv. Chem*. ID: 834641 (2014).
21. Bakavoli M., Beyzaei H., Rahimizadeh M. and Eshghi H. Regioselective synthesis of new 2-(E)-cyano(oxazolidin-2-ylidene)thiazoles. *Eur. J. Chem*. **2** (3): 356-358 (2011).
22. Bakavoli M., Beyzaei H., Rahimizadeh M., Eshghi H. and Takjoo R. Regioselective synthesis of new 2-(E)-cyano(thiazolidin-2-ylidene)thiazoles. *Molecules* **14**: 4849-4857 (2009).
23. Bakavoli M., Beyzaei H., Rahimizadeh M. and Eshghi H. Regioselective synthesis of 2[(E)-(benzo[*d*]thiazol-2(3H)ylidene)(cyano)methyl]thiazoles. *Heterocycl. Commun*. **17** (3-4): 151-154 (2011).
24. Bakavoli M., Beyzaei H., Rahimizadeh M. and Eshghi H. One-pot synthesis of functionalized tetrahydro-1,4-thiazepines. *Synth. Commun*. **41** (8): 1181-1185 (2011).
25. Juspin T., Laget M., Terme T., Azas N. and Vanelle P. TDAE-assisted synthesis of new imidazo[2,1-b]thiazole derivatives as anti-infectious agents. *Eur. J. Med. Chem*. **45** (2): 840-845 (2010).
26. Ghasemi B., Sanjarani G., Sanjarani Z. and Majidiani H. Evaluation of anti-bacterial effects of some novel thiazole and imidazole derivatives against some pathogenic bacteria. *Iran. J. Microbiol.* **7** (5): 281-286 (2015).
27. Brvar M., Perdih A., Oblak M., Masic L.P. and Solmajer T. In silico discovery of 2-amino-4-(2,4-dihydroxyphenyl)thiazoles as novel inhibitors of DNA gyrase B. *Bioorg. Med. Chem. Lett.* **20** (3): 958-962 (2010).
28. Lu X., Liu X., Wan B., Franzblau S. G., Chen L. and Zhou C. Synthesis and evaluation of anti-tubercular and antibacterial activities of new 4-(2,6-dichlorobenzyloxy)phenyl thiazole, oxazole and imidazole

derivatives. *Eur. J. Med. Chem*. **49**: 164-171 (2012).
29. Bartels H. and Bartels R. Simple, rapid and sensitive determination of protionamide in human serum by high-performance liquid chromatography. *J. Chrom. B Biomed. Sci. Appl.* **707** (1-2): 338-341 (1998).
30. Xue T., Ding S., Guo B., Chu W., Wang H. and Yang Y. Synthesis and structure–activity relationship studies of novel [6, 6, 5] tricyclic oxazolidinone derivatives as potential antibacterial agents. *Bioorg. Med. Chem. Lett*. **25** (10): 2203-2210 (2015).
31. Rajkumar R., Kamaraj A. and Krishnasamy K. Synthesis, spectral characterization and biological evaluation of novel 1-(2-(4, 5-dimethyl-2-phenyl-1H-imidazol-1-yl) ethyl) piperazine derivatives. *J. Saudi Chem. Soc*. **18** (5): 735-743 (2014).
32. Wang S.F., Yin Y., Wu X., Qiao F., Sha S. and Lv P.C. Synthesis, molecular docking and biological evaluation of coumarin derivatives containing piperazine skeleton as potential antibacterial agents. *Bioorg. Med. Chem*. **22** (21): 5727-5737 (2014).
33. Madabhushi S., Mallu K. K., Vangipuram V. S., Kurva S., Poornachandra Y. and Ganesh Kumar C. Synthesis of novel benzimidazole functionalized chiral thioureas and evaluation of their antibacterial and anticancer activities. *Bioorg. Med. Chem. Lett*. **24** (20): 4822-4825 (2014).
34. Elumalai K., Ashraf Ali M., Elumalai M., Eluri K. and Srinivasan S. Novel isoniazid cyclocondensed 1,2,3,4-tetrahydropyrimidine derivatives for treating infectious disease: a synthesis and *in vitro* biological evaluation. *J. Acute Dis*. **2** (4): 316-321 (2013).
35. Akhaja T. N. and Raval J. P. Design, synthesis, in vitro evaluation of tetrahydropyrimidine-isatin hybrids as potential antibacterial, antifungal and anti-tubercular agents. *Chin. Chem. Lett*. **23** (4): 446-449 (2012).
36. Kumar M., Sharma K., Kumar Fogla A., Sharma K. and Rathore M. Synthesis and antimicrobial activity of 2,4-diaryl-2,3-dihydrobenzo[b][1,4]thiazepines. *Res. Chem. Intermediat.* **39** (6): 2555-2564 (2013).