Synthesis, antibacterial and QSAR evaluation of 5-oxo and 5-thio derivatives of 1,4-disubstituted tetrazoles

Rami Y. Morjan, Nisreen H. El-Attar, Omar S. Abu- Teim, Maschke Ulrich, Adel M. Awadallah, Ahmed M. Mkadmh, Abdelrauof A. Elmanama, James Raftery, Fakhr M. Abu-Awwad, Zyad J. Yaseen, Ammar F. Elqidrea, John M. Gardiner

PII: S0960-894X(15)00402-3
DOI: http://dx.doi.org/10.1016/j.bmcl.2015.04.070
Reference: BMCL 22653

To appear in: *Bioorganic & Medicinal Chemistry Letters*

Received Date: 10 March 2015
Revised Date: 19 April 2015
Accepted Date: 20 April 2015

Please cite this article as: Morjan, R.Y., El-Attar, N.H., Abu- Teim, O.S., Ulrich, M., Awadallah, A.M., Mkadmh, A.M., Elmanama, A.A., Raftery, J., Abu-Awwad, F.M., Yaseen, Z.J., Elqidrea, A.F., Gardiner, J.M., Synthesis, antibacterial and QSAR evaluation of 5-oxo and 5-thio derivatives of 1,4-disubstituted tetrazoles, *Bioorganic & Medicinal Chemistry Letters* (2015), doi: http://dx.doi.org/10.1016/j.bmcl.2015.04.070

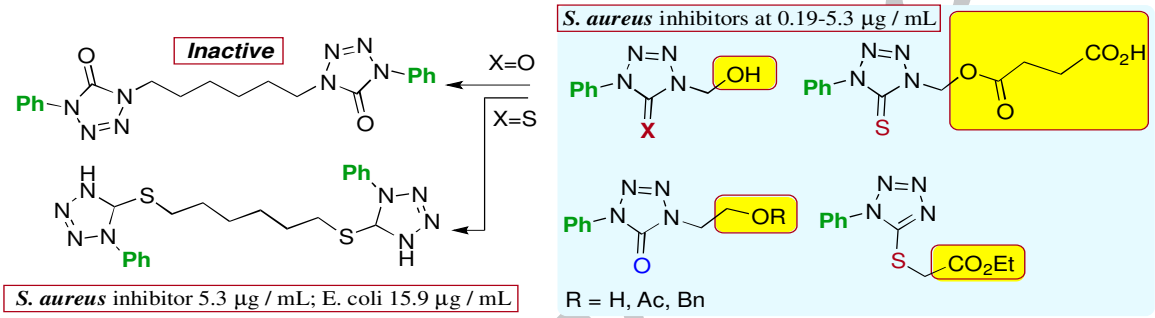
Graphical Abstract

To create your abstract, type over the instructions in the template box below.
Fonts or abstract dimensions should not be changed or altered.

Synthesis, antibacterial, and QSAR evaluation of 5-oxo and 5-thio derivatives of 1,4-disubstituted tetrazoles

Leave this area blank for abstract info.

Rami Y. Morjan, Nisreen H. El-Attar, Omar S. Abu-Teim, Maschke Ulrich, Adel M. Awadallah, Ahmed M. Mkadmh, Abdelrauof A. Elmanama, James Raftery, Fakhr M. Abu-Awwad, Zyad J. Yaseen, Ammar F. Elqidra, and John M. Gardiner*



Bioorganic & Medicinal Chemistry Letters
journal homepage: www.elsevier.com

Synthesis, antibacterial and QSAR evaluation of 5-oxo and 5-thio derivatives of 1,4-disubstituted tetrazoles

Rami Y. Morjan[a], Nisreen H. El-Attar[b], Omar S. Abu- Teim[b], Maschke Ulrich[c], Adel M. Awadallah[a], Ahmed M. Mkadmh[d], Abdelrauof A. Elmanama[e], James Raftery[f], Fakhr M. Abu-Awwad[a], Zyad J. Yaseen[a], Ammar F. Elqidrea[c] and John M. Gardiner[g,*]

[a]*Chemistry Department, Faculty of Science, Islamic University of Gaza, PO Box 108, Gaza Strip, Palestine.*
[b]*Chemistry Department, Faculty of Science, Al-Azhar University, P. O. Box 1277, Gaza, Palestine.*
[c]*UMET, UMR CNRS N°8207, Université Lille 1 - Sciences et Technologies, 59655 Villeneuve d'Ascq Cedex, France.*
[d]*Chemistry Department, Faculty of Applied Science, Alaqsa University, PO Box 4051, Gaza, Palestine.*
[e]*Medical Laboratory Sciences Department, Faculty of Health Science, Islamic University of Gaza, P.O. Box 108, Gaza Strip, Palestine.*
[f]*School of Chemistry, The University of Manchester, Brunswick Street, Manchester, M13 9PL, United Kingdom.*
[g]*Manchester Institute of Biotechnology, School of Chemistry and EPS, The University of Manchester, Manchester M1 7DN, United Kingdom.*

ARTICLE INFO

Article history:
Received
Revised
Accepted
Available online

Keywords:
Tetrazole
Tetrazolones
Tetrazolethiones
Antibacterial
QSAR

ABSTRACT

A series of 1,4-disubstituted tetrazol-5-ones **3a**, **5**, **7**, **12**, **13** 1,4-disubstituted tetrazol-5-thiones **3b**, **9**, **10** was synthesized and fully characterized by IR, MS, ^{1}H NMR and ^{13}C NMR. The series was evaluated for *in vitro* antibacterial activity against four Gram negative (*Escherichia coli, Proteus mirabilis, Klebsiella pneumonia, and Pseudomonas aeruginosa*) and three Gram positive (*Staphylococcus aureus, Enterococcus faecalis, and Bacillus subtilis*) bacteria. The zone of inhibition was measured using the well-diffusion assay, and *in vitro* minimum inhibitory concentration (MIC) was determined by microbroth dilution assay. MIC values indicate that compounds exhibited a varied range (0.2–37 μg/mL) of antibacterial activity against the tested bacterial strains. Statistically significant QSAR models were developed by the simple linear regression analysis for the correlation of MIC with computed descriptors. The concluded cross validated regression factors are 0.953 and 0.986 for *E. coli*, and *S. aureus* respectively.

Microbial infections have been of recent concern due to the increasing emergence of multidrug-resistant strains, intractable pathogenic microorganisms and newly arising pathogens[1–4]. Various synthetic and semi-synthetic antimicrobial agents have been discovered and extensively used in the clinic to treat various community, environment and hospital-acquired microbial infections[5-6]. There is a continuous demand to find drugs that have effects on Gram-positive and Gram-negative, as well as multidrug-resistant bacteria[7-8]. However, and despite significant progress in this field; there are still some unresolved problems for development of new clinical drugs, including narrow antimicrobial spectrum, adverse effects and high toxicity[9].

The discovery and development of structurally-novel antimicrobial agents with good pharmacological profile and excellent activity towards resistant strains are highly desirable[10–11]. Tetrazoles have received attention in recent years due to widespread applications notably in the field of medicinal chemistry due to their privileged biological activity[12-13]. Many tetrazoles possess hypotensive[14], antimicrobial[15], antiviral[16], antiallergic[17], cytostatic[18], antifungal activity[19]. The tetrazole ring is also an isostere of carboxyl[20-21], amide[22], imidazole[23], benzimidazole[24] and carbazole groups[25] in designing various new types of drug molecules. Dozens of highly effective drugs whose active pharmaceutical ingredients contain the tetrazole ring are reported and approved by FDA. Examples are Losartan, Valsartan, Irbesartan, Flomoxef and Cefonicid[26-30]. Various methodologies for preparing compounds with a tetrazole ring system have been developed, among the most important are those based on cycloaddition reaction of azides with cyanide or isonitriles[31-33]. Tetrazolones and tetrazolethiones have also received considerable attention in the last few years. Many pesticides[34] and herbicides[35], and drugs have been patented for the treatment of central nervous system disorders,[36] HIV,[37] sexual dysfunction,[38] asthma, [39] diabetes, [40] and also a number of antiviral[41], antibacterial[42] antimicrobial[43], and anti-inflammatory[44] agents have been reported containing one of

* Corresponding author. Tel.: +44-161-306-4530; e-mail: gardiner@manchester.ac.uk

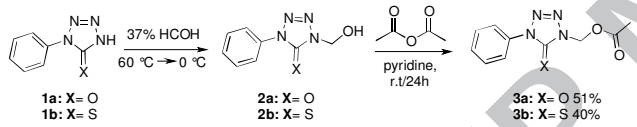
these ring systems. In a local study conducted at Al-shifa hospital`s burn unit it was shown that *Pseudomonas* isolates were resistant to most of antimicrobials used except for piperacillin-tazobactam. The incidence of methicillin-resistant Staphylococci according to the oxacillin sensitivity test was 60% in patient samples, 77.8% in health care worker samples and 90% in environmental samples[45]. This indicates the necessity for new active compounds that could supercede old inactive antimicrobials.

Quantitative structure activity relationship (QSAR) is an important method employed for the prediction of biological activity of compounds and in finding quantitative correlations between the molecular structures and their biological activities [46].

Herein, we describe the synthesis, antimicrobial activity and QSAR investigations for a series of novel 1,4-disubstituted tetrazol-5-one-5-thione.

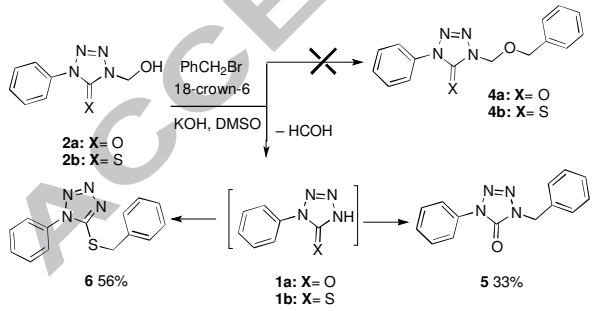
The starting materials, 1,4-dihydro-1-hydroxymethyl-4-phenyl tetrazol-5-one **2a** and its thione counterpart **2b**, were synthesized according to the reported procedures.[47] These were intended to be substrates for the synthesis of a range of *O*-alkylated and *O*-acylated derivatives for biological evaluation.

Initial work involved targeting acylation and benzylations. Reaction of either **2a** or **2b** with acetic anhydride in pyridine afforded the novel compounds **3a** and **3b** in 51% and 40% yield, respectively (**Scheme 1**).



Scheme 1: Reaction of **2a** and **2b** with acetic anhydride.

By contrast, reaction of **2a** or **2b** with benzyl bromide using KOH and 18-crown-6 as phase transfer catalyst in DMSO, at room temperature, afforded compounds **5** and **6** in 33% and 56% yield respectively, instead of the expected products **4a** and **4b** (**Scheme 2**).



Scheme 2: Reaction of tetrazolinone **2a** and **2b** with benzyl bromide.

The synthesis of compound **5** was previously reported *via* direct reaction of the parent tetrazole **1a** with benzyl bromide [48] and compound **6** has been prepared directly *via* thiol alkylation at sulfur[49]. The formation of compounds **5** and **6** in the reactions of Scheme 2 is believed to arise *via in situ* loss of formaldehyde (HCOH) from the starting materials **2a** and **2b** to form the NH tetrazoles **1a** and **1b**, respectively, which then undergo direct alkylation at N4 of the tetrazole ring of **1a**. In the case of **1b** the alkylation occurs at S rather than at N as a result of thione/thiol tautomerism leading to the formation of **6**.

We obtained crystals of *N*-benzyl derivative **5** and obtained an X-ray crystal structure[50]. The crystal structure of compound **5** has a space point group P 21/n with a unit cell dimensions 8.0027(8) Å, 13.0948(11) Å, 12.0557 Å and cell angles 90.00, 105.462(7), and 90.00 for α, β and γ, respectively (**Figure 1**). The crystal packing diagram and full details of the crystal structure are shown in (**Figure S1, Supplementary Data**)

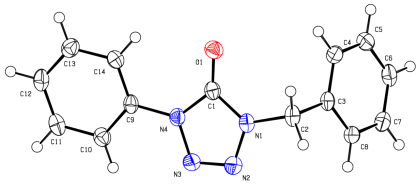
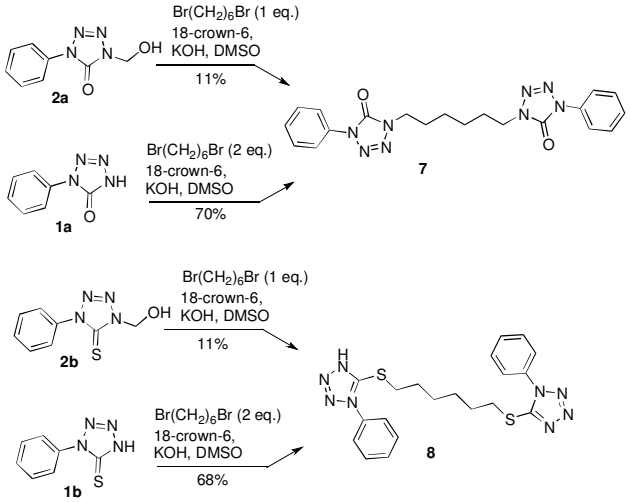


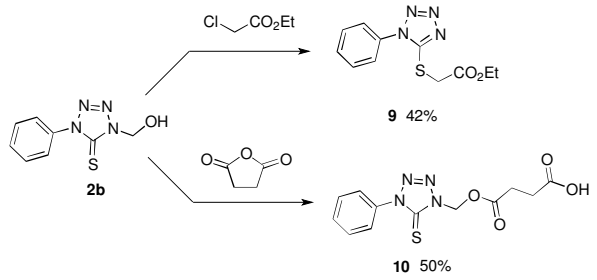
Figure 1. X-ray structure of **5**

Following from the outcomes of acylation and benzylation reactions, we evaluated reactions of **2** with a bis-alkylating agent. The reaction of **2a** and **2b** with one equivalent of 1,6-dibromohexane afforded the bis-tetrazole dimeric compounds **7** and **8** in 11% and 10% yield, respectively. The yields were significantly increased when two equivalents of **1a** and **1b** were reacted with one equivalent of the dibromohexane as this produced **7** and **8** in 70% and 68% respectively (**Scheme 3**). Compound **8** was also previously reported *via* reaction of two equivalents of 1,6-dibromohexane with 5-mercapto-1-phenyltetrazole[55]. The outcomes from **2a** and **2b** are attributed again to the *in situ* deformylation to generate **1a** and **1b** followed by direct reaction with 1,6-dibromohexane to form **7** and **8**, however, this does not appear to be as effective as direct N-alkylations for the dimerizations.



Scheme 3: Synthesis of dimers **7** and **8**.

Alkylation at S following *in situ* loss of formaldehyde from **2b** was also observed using ethyl 2-chloroacetate as the electrophile, thereby giving rise to the known[56] compound **9** as the only product (**Scheme 4**).

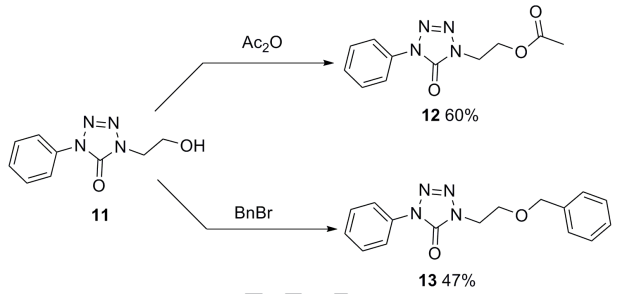


Scheme 4: Reactions of **2b** with ethyl 2-chloroacetate or succinic anhydride.

Similarly to acylation (cf. Scheme 1), reaction of **2b** with succinic anhydride afforded the expected compound **10** in 50% yield. No evidence of any by-product formation as a result of losing formaldehyde was observed. In order to avoid the *in situ* formation of compounds **1a** and **1b** and to be able to extend the linkage at N4 of the tetrazol-5-one/thione rings, we employed a different starting material, namely the homologous hydroxyethyl-bearing **11**. The reaction of **11** with acetic anhydride afforded the acylation product **12** in 60% yield, but the key difference was that *O*-alkylations now proceeded without the *N*-alkylation seen with **2a**. The reaction of **11** with benzyl bromide under phase transfer catalyst conditions afforded compound **13** in 47% (**Scheme 5**).

Scheme 5: Reaction of **11** with acetic anhydride and benzyl bromide

With this series of six new tetrazoles along with the known compounds prepared, we undertook evaluation of their antibacterial effects. The well-diffusion assay methodology[57] was employed to screen DMSO solutions of 1,4-disubstituted tetrazol-5-ones **3a**, **5**, **7**, **12**, **13** and 1,4-disubstituted tetrazol-5-



thiones **3b**, **9**, **10**, and the starting materials **1a**, **1b**, **2a** and **2b** for antimicrobial activity. Four Gram negative (*Escherichia coli*, *Proteus mirabilis*, *Klebsiella pneumonia*, and *Pseudomonas aeruginosa*) and three Gram positive isolates (*Staphylococcus aureus*, *Enterococcus faecalis*, and *Bacillus subtilis*) obtained from Al-Shifa Hospital Microbiology laboratory were used as test organisms. All compounds either in powder or oil forms were weighed (range from 45 to 250 mg) and dissolved in DMSO. The volume of DMSO used ranged from 750 to 2000 µl. In short, 6 holes were punched on each of three Muller Hinton agar plates that were inoculated with standardized bacterial suspension [58]. 100 µl of each of the tested compound solutions were carefully dispensed in the corresponding hole. Plates were set for 30 minutes at the refrigerator temperature and then incubated for 24 hours at 37 ºC. The zone of inhibition in mm for the tested compounds is provided in **Table 1**.

All compounds that showed measurable antibacterial activity were further tested to determine their minimum inhibitory concentration (MIC) using microbroth dilution assay. Each compound was serially diluted in fixed volumes containing standardized bacterial suspension (standardization was made by comparing growth to 0.5 McFarland turbidity standards). A positive growth control was included for each plate.

Table 1: Zone of inhibition in mm for compounds tested[a]

Zone of inhibition in mm							
	Gram-positive bacteria			Gram-negative bacteria			
Compound	***S. a***	***S.s***	***B. s***	***E.c***	***K. p***	***P. m***	***P.a***
1a	12	11	14	10	9	0	0
1b	18	12	16	14	8	19	11
2a	38	42	33	30	22	34	24
2b	32	42	30	31	19	26	23
3a	9	10	14	0	0	11	0
3b	10	0	12	0	0	15	0
5	0	0	0	0	0	7	0
6	0	0	0	0	0	0	0
7	0	0	0	0	0	0	0
8	0	0	0	0	0	0	0
9	9	13	13	0	0	14	0
10	24	19	12	11	16	11	17
11	0	0	12	11	13	12	0
12	0	0	9	12	0	15	0
13	0	0	0	0	0	20	0

[a] *S. a Staphylococcus aureus,* S.s *Streptococcus spp., B.s Bacillus subtilis, E.c* Escherichia coli, *K.p, Klebsiella pneumonia P.m, Proteus mirabilis, P.a Pseudomonas aeruginosa*.

Microtiter plates were incubated for 24 hours at 37 ºC. Growth inhibition was detected by adding 20 µl of 0.5% aqueous 2,3,5-triphenyltetrazolium chloride (TTC), supplied by Merck. MIC was defined as the lowest concentration of inoculum that inhibited visible growth, as indicated by TTC color change (red to colorless)[59].

Table 2. The MIC results of compounds that exhibited antibacterial activity (µg / mL)

Compound	***E. coli***	***S. aureus***
1a	1.5432	1.5432
1b	1.7672	1.7637
2a	1.76	0.19
2b	0.58	1.76
3a	13.88	2.66
3b	13.88	4.62
9	15.87	5.29
10	1.37	1.37
11	8.00	5.29
12	37.03	37.03

Compounds **1a**, **1b**, **2a, 2b, 3a, 3b** and **9-12** were evaluated for their antibacterial activity against Gram positive (*Staphylococcus aureus)* and Gram negative (*Escherichia coli*) bacteria to determine the MIC. These compounds showed antibacterial activities by the microbroth dilution assay and the MIC results are summarized in **Table 2**. Zones of inhibition around the ten compounds (**1a**, **1b**, **2a, 2b, 3a, 3b, 9-12**) by the microbroth dilution method are shown in (**Figure S2**, **Supplementary Data)**. Reduction of 2,3,5-triphenyltetrazolium chloride giving a red color was used as an indicator of bacterial growth in MIC determination (**Figure S3, Supplementary Data**).

A notable broad-spectrum effect of some of the tested compounds was evident, especially, for compounds **2a**, **2b**, and

10, whilst **3a**, **10** and **11** all showed activity against 4 of the panel of microrganisms. In addition, the effects on *P. aeruginosa* of **2a**, **2b**, and **10** are particularly promising. MIC values indicate that the ten compounds reported in Table 2 exhibited a varied range (0.2–37 μg/mL) of antibacterial activity. The MIC values of compounds **2a** and **2b** showed the maximum inhibition activity (0.19 and 0.59 mg/mL respectively). Further studies to determine lethal dose, safety and mechanism of action of these compounds will be our priority with the results reported here showing that readily-accessible 1,4-differentially-substituted tetrazoles – the chemistry herein illustrating the capacity for further structural diversification - offer a promising scaffold for new antibacterial discovery.

Computational and QSAR Work

The molecular geometries of the structures listed in **Table 2** were fully optimized at B3LYP/6-31G(d) level of theory using Gaussian 2003 for windows (G03W) without any applied molecular symmetry constraint[60,61]. The optimized structures were properly attributed to their local minima with no imaginary frequencies, followed by QSAR calculations at the same level of theory using CODESSA software (version 2.10)[62].

In QSAR procedures, 300 physicochemical descriptors were computed for each of the optimized structures. Employing a heuristic method for the whole dataset, the validated descriptors are then ranked according to their correlation coefficients.

In **Figure 2(a, b)**, the QSAR-based correlations between the experimental MIT in **Table 2** and the computed MIT according to the reached models are depicted. Four statistical parameters are employed to verify the findings. The squared correlation coefficient (R^2), The squared cross-validated correlation coefficient (R^2_{cv}), Fisher F-criterion value, and the squared standard deviation of the regression (s^2). For *Escherichia coli* bacteria, the reached correlation in **Figure 2a** was based on three descriptors; relative number of H atoms, HASA-2/SQRT(TMSA) [Zefirov's PC], and HA dependent HDCA-2/TMSA [Quantum-Chemical PC] with R^2 = 0.987, R^2_{CV} = 0.953, F = 150.62, s^2 = 2.55.

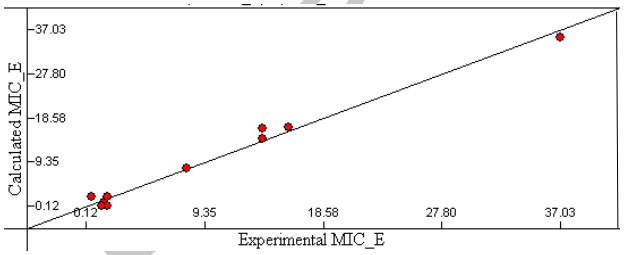


Figure 2a

Similarly, in **Figure 2b** for *Staphylococcus aureus* shows a linear correlation with R^2 = 0.999, R^2_{CV} = 0.986, F = 807.03, s^2 = 0.34. The four descriptors included in the model were relative number of H atoms, HASA-2 [Zefirov's PC], FPSA-3 Fractional PPSA (PPSA-3/TMSA) [Zefirov's PC], and HA dependent HDSA-1/TMSA [Zefirov's PC].

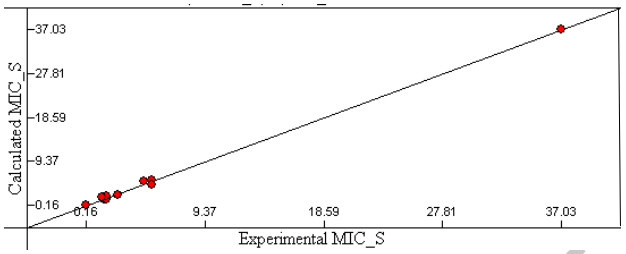


Figure 2b

Figure 2. Correlations between experimental and calculated MIC values (**2a**): *Escherichia coli* (**2b**): *Staphylococcus aureus*.

Acknowledgments

The authors would like to thank the French Government for the financial support of this project through Al Maqdisi l2Fl program

Supplementary Material

Supplementary data (representative experimental procedures, characterization data, and copies of spectra, and images of assays) associated with this article can be found, in the online version, at http://….

References

1) Scheffler, R. J.; Colmer, S.; Tynan, H.; Demain, A. L.; Gullo, V. P. *Appl. Microbiol. Biotechnol.* **2013,** *97*, 969.
2) Zhang, L.; Peng, X. M.; Damu, G. L. V.; Geng, R. X.; Zhou, C. H. *Med. Res. Rev.* **2014,** *34*, 340.
3) Peng, X. M.; Cai, G. X.; Zhou, C. H. *Curr. Top. Med. Chem.* **2013**, *13*, 1963.
4) Peng, X. M.; Damu, G. L. V.; Zhou, C. H. *Curr. Pharm. Des*. **2013**, *19*, 3884.
5) Davies, J.; Davies, D. *Microbiol. Mol. Biol. Rev.* **2010**, *74*, 417
6) Genin, M. J.; Allwine, D. A.; Anderson,D. J.; Barbachyn, M. R.; Emmert, D. E.; Garmon, S. A.; Graber, D. R.; Grega, K. C.; Hester, J. B.; Hutchinson, D. K.; Morris, J.; Reischer, R. J.; Ford, C. W.; Zurenko, G. E.; Hamel, J. C.; Schaadt, R. D.; Stapert, D.; Yagi, B. H. *J. Med. Chem.* **2000***, 43,* 953.
7) Hokai, Y.; Jurkowicz, B.; Fern, J.; Fernández-Gallardo, J.; Zakirkhodjaev, N.; Sanaú, M.; Muth, T. R.; Contel, M. *J. Inorg. Biochem.* **2014**, *138*, 81.
8) Cassetta, I. M.; Marzo, T.; S. Fallani, Novelli, A.; Messori, L. *Biometals* **2014**, *27*, 787.
9) Dai, L.-L.; Zhang, H.-Z.; Nagarajan, S.; Rasheed, S.; Zhou, C.-H. *Med. Chem. Commun*. **2015**, DOI: 10.1039/c4md00266k
10) Yin, B. T.; Yan, C. Y.; Peng, X. M.; Zhang, S. L.; Rasheed, S; Geng, R. X.; Zhou, C. H. *Eur. J. Med. Chem.* **2014**, *71*, 148.
11) Cui, S. F.; Ren,Y.; Zhang, S. L.; Peng, X. M.; Damu, G. L. V.; Geng, R. X.; Zhou, C. H. *Bioorg. Med. Chem. Lett*. **2013**, *23*, 3267.
12) Ostrovskii, V. A.; Trifonov, R. E.; Popova, E. A. *Russ. Chem. Bull. Int. Ed.* **2012***, 61,* 768.
13) Singh, H.; Chawla, A. S.; Kapoor, V. K.; Paul, D.; Malhotra, R. K. *Progress in Medicinal Chemistry* **1980**, *17*, 151.
14) Bourdonnec, B. Le.; Bourdonnec, Meulon, E.; Yous, S.; Goossens, J. F.; Houssin, R.; Hénichart, J. P. *J. Med. Chem.* **2000**, *43*, 2685.
15) Tremblay, L. W.; Xu, H.; Blanchard, J. S. *Biochemistry* **2010**, *49*, 9685.
16) Diwakar, S. D.; Bhagwat, S. S.; Shingare, M. S.; Gill, C. H. *Bioorg. Med. Chem. Lett.* **2008**, *18*, 4678.

17) A. Rajasekaran, P. P. Thampi, *Eur. J. Med. Chem*. **2004**, *39*, 273.

18) Kumar, Ch. N. S. S. P.; Parida, D. K; Santhoshi, A.; Kota, A. K.; Sridhar, B.; Rao, V. J. *Med. Chem. Commun*. **2011**, *2*, 486.

19) Ichikawa,T.;, Kitazaki, T.; Matsushita, Y.; Yamada, M.; Hayashi, R.; Yamaguchi, M.; Kiyota, Y.; Okonogi, Itoh, K. *Chem. Pharm. Bull*. **2001**, *49*, 1102.

20) Pegklidou, K.; Koukoulitsa, C.; Nicolaou, I.; Demopoulos, V. J. *Bioorg. Med.Chem*. **2010**, *18*, 2107.

21) Matta, C. F.; Arabi, A. A.; Weaver, D. F. *Eur. J. Med. Chem.* **2010**, *45*, 1868.

22) Al-Hourani, B. J.; Sharma, S. K.; Mane, J. Y.; Tuszynski, J.; Baracos, V.; Kniess, T.; Suresh, M.; Pietzsch, J.; Wuest, F. *Bioorg. Med. Chem. Lett*. **2011**, *21*, 1823.

23) Carroll, W. A.; Kalvin, D. M.; Medrano, A. P.; Florjancic, A. S.; Wang, Y.; Donnelly-Roberts, D. L.; Namovic, M. T.; Grayson, G.; Honoréa, P.; Jarvis, M. F. *Bioorg. Med. Chem. Lett*. **2007**, *17*, 4044.

24) Shi, Y.; Zhou, C. H.; Zhou, X. D.; Geng, R. X.; Ji, Q. G. *Acta Pharmacol. Sin*. **2011**, *46*, 798.

25) Zhang, F. F.; Gan, L. L.; Zhou, C. H. *Bioorg. Med. Chem. Lett*. **2010**, *20*, 1881.

26) MacFadyen, R. J.; Tree, M.; Lever, A. F.; Reid, J. L. *Clin. Sci*. **1992**, *83*, 549.

27) Karaman, M.; Balta, S.; Ay, S. A.; Cakar, M.; Naharci, I.; Demirkol, S.; Celik, T.; Arslan, Z.; Kurt, O.; Kocak, N.; Sarlak, H.; Demirbas, S.; Bulucu F.; Bozoglu, E. *Clin. Exp. Hypertens.* **2013**, *35*, 516.

28) Bauer, M.; Harris, R. K.; Rao, R. C.; Apperley, D. C.; Rodger, C. A. *J. Chem. Soc., Perkin Trans*. **1998**, *2*, 475.

29) Lee, C. H.; Liu, J. W.; Li, C. C.; Tang, Y. F.; Su, L. H. *Antimicrob. Agents Chemother*. **2011**, *55*, 4058.

30) Pochini, L.; Galluccio, M.; Scumaci, D.; Giangregorio, N.; Tonazzi, A.; Palmieri, F.; Indiveri, C. *Chem. Bio .Interact*. **2008**, *173*, 187.

31) Vandensavel, J. M.; Smets, G.; L'Abbe, G. *J. Org. Chem*. **1973**, 38, 675.

32) Lieber, E.; Ramachandran, J. *Can. J. Chem.* **1959**, *37*, 101.

33) Horwitz J. P.; Fisher, B. E.; Tomasewski A. *J. Am. Chem. Soc*. **1959**, *81*, 3076.

34) Luo, Y.-P.; Yang, G.-F. *Bioorg. Med. Chem*. **2007**, *15*, 1716.

35) Foldenyi, R. *Monatsh. Chem.* **1995**, *126*, 1035.

36) Andres-Gil, J. I.; Alcazar-Vaca, M. J.; Pastor-Fernandez, J.; Drinkenburg, W. H. I. M.; Langlois, X. J. M.; Oyarzabal-Santamarina, J.; Vega-Ramiro, J. A. EP 569512006067139, 2006.

37) Show-Reid, C. A.; Miller, M. D.; Hazuda, D. J.; Ferrer, M.; Sur, S. M.; Summa, V.; Lyle, T. A.; Kinzel, O.; Pescatore, G.; Muraglia, E.; Orvieto, F.; Williams, P. D. U.S. 16671 2005115147, 2005.

38) Cirino, G.; Mirone, V.; Ignarro, L. J.; Del Soldato, P. EP 50779 2004043443, 2004.

39) Janssens, F. E.; Sommen, F. M.; Surleraux, D. L. N. G.; Leenaerts, J. E.; Van Roosbroeck, Y. E. M. EP 4660 9716440, 1997.

40) Shih, T. L.; Candelore, M. R.; Cascieri, M. A.; Chiu, S.-H. L.; Colwell, L. F., Jr.; Deng, L.; Feeney, W. P.; Forrest, M. J.; Hom, G. J.; MacIntyre, D. E. *Bioorg. Med. Chem. Lett*. **1999**, *9*, 1251.

41) Shenk, T.; Rabinowitz, J. D.; Munger, J.; Bennett, B. U.S. 6959 2009023059, 2009.

42) Kang, H. Y.; Koh, H. Y.; Koh, S. H.; Chang, M. H. *Korean J. Med. Chem.* **1991**, *1*, 18.

43) Sindkhedkar, M. D.; Bhavsar, S. B.; Patil, V. J.; Deshpande, P. K.; Patel, M. V. IN 208 2007023507, 2007.

44) Dunlap, R. P.; Boaz, N. W.; Mura, A. J.; Hlasta, D. J. U.S. 2434 9013549, 1990.

45) Elmanama, A.; Laham, N.; Tayh, G. *Burns* **2013**, *39*, 1612.

46) Verma, J.; Khedkar, M. V.; Coutinho C. E. *Current Topics in Medicinal Chemistry* **2010**, *10*, 95.

47) Awadallah, A.; Rademacher, P.; Boese, R. *J. Prakt. Chem*. **1995**, *337*, 636.

48) Poplavskaya, V. Yu.; Alam, L. V.; Koldobskii, G. I. *Russ. J. Org. Chem.* **1999**, *36*, 1793.

49) Savolainen, A. M.; Han, X.; Wu, J. *Org. Lett*. **2014,** *16,* 4349.

50) Cambridge crystallographic data centre for small molecules CCDC for compound **5** (1035478).

51) Bondi, A. *J. Phys. Chem*. **1964**, *68*, 441.

52) Rowland, R. S.; Taylor, R. *J. Phys. Chem*. **1996**, *100*, 7384.

53) Becke, A. D. *J. Chem. Phys*. **1993**, *98*, 5648.

54) Lee, C.; Yang, W.; Parr, R. G. *Phys. Rev. B*, **1988**, *37*, 785.

55) He, D.-P.; Dou, Y.-L.; She, J.-B. *Z. Kristallogr*, **2006**, *221*, 25.

56) R. Neidlein, J. Tauber, Chem. Ber., **1967**, 100, 736-740

57) Valgas, C.; de Souza, S. M.; Smânia, F. A. Elza.; Smânia Jr. *Brazilian Journal of Microbiology*, **2007**, *38*, 369.

58) Elmanama, A. A.; Alyazji, A. A.; Ganema. E.A. *Ann Alquds Med* **2011**, *7*, 33.

59) Morjan, R. Y.; Mkadmh, A. M.; Beadham, I.; Elmanama, A. A.; Mattar, M. R.; Raftery, J.; Pritchard, R. G.; Adel M. Awadallah, A. M.; Gardiner, J. M. *Bioorg. Med. Chem. Lett*. **2014**, *24*, 5796.

60) Dewar, M. J.S.; Zoebisch, E.G.; Healy, E.F.; Stewart, J.J.P.; *JACS* **1985,** 107, 3902.

61) Frisch, M. J., (1995) Gaussian (03W) **2003**, Gaussian, Inc., Pittsburgh, PA, USA.

62) CODESSATM, V.2.7.10 (PC), Semichem, 7204 Mullen, Shawnee, KS, USA, Copyright Semichem and the University of Florida, **2002.**