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Synthesis and antimicrobial activity of some new macrocyclic *bis*-sulfonamide and disulfides

Hossein Eshghi[a,*], Mohammad Rahimizadeh[a], Mahmood Zokaei[b], Shaghayegh Eshghi[c], Shohreh Eshghi[c], Zinab Faghihi[a], Elaheh Tabasi[b] and Mehdi Kihanyan[a]

[a] *Department of Chemistry, School of Sciences, Ferdowsi University of Mashhad, Mashhad, IR-91775-1436, Iran*
[b] *Department of Biology, School of Sciences, Ferdowsi University of Mashhad, Mashhad, IR-91775-1436, Iran*
[c] *School of Medicine, Mashhad University of Medical Sciences, Mashhad, IR-91779-48564, Iran*

**Corresponding author at: Department of Chemistry, School of Sciences, Ferdowsi University of Mashhad, Mashhad, IR-91775-1436, Iran. Tel.: +98.511.8795457; fax: +98.511.8795457. E-mail address: heshghi@ferdowsi.um.ac.ir (H. Eshghi).*

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ABSTRACT

Synthesis and antimicrobial evaluation of some typical macrocyclic crown ethers including amide, sulfonamide, and disulfide moieties are reported. Novel macrocyclic *bis*-sulfonamide, amide, and disulfides are prepared by reacting the *bis*-chlorides and diamines by fast addition method. The antimicrobial activities of the synthesized compounds are measured. *Bis*-sulfonamide and disulfide crown ethers showed antibacterial activities against most strains tested.

1. Introduction

There have been great advances in the chemistry of sulfur-substituted crown ethers [1]. Application of these compounds to molecular machines and devices has also attracted much attention [2]. In general, redox reactions between dithiol and disulfide are very useful to control molecular structures and act as function simultaneously [3]. This phenomenon is known to regulate enzymatic activity and ion recognition of artificial hosts [4]. Ryser and co-workers have shown for the first time that agents which interfere with thiol-disulfide reduction on the cell surface inhibit HIV-1 infection. They identified protein disulfide isomerase (PDI) located on the cell surface as a candidate enzyme to catalyze this reaction [5]. Accordingly, design and synthesis of several redox-switched crown ethers with disulfide linkage are reported, for examples, for zinc abstraction from proteins and disrupt their conformation for achieving maturation of HIV virus [6], paraquat and secondary ammonium salts recognitions [7], and selective electrode properties in laboratory [8], and in the biomemberanes [9].

On the other hand, the sulfonamide functional group has a long and rich history in organic chemistry and drug discovery. Beginning with the discovery of the 'sulfa' antibiotics in the 1930s that revolutionized the treatment of bacterial infections [10], and continuing into the present day with the development of potent anti-retrovirals used to treat patients infected with HIV [11], sulfonamides remain a particularly important class of compounds for the treatment of infectious diseases [12].

Recently, there has been much interest in the chemistry of proton-ionizable crown ethers [13,14]. The nature of the proton-ionizable group, particularly its acidity, controls the metal ion complexation properties of such ligands. Aryl sulfonamides offer an advantage over other proton-ionizable crown compounds by increasing the acidity of the amide protons by stabilizing the nitrogen anions [15,16]. Bradshaw *et al.* [15-17] have been reported *bis*-sulfonamide crown compounds as excellent alkali metal cation carriers in bulk liquid membranes.

In recent years, we have been involved in the use of macrocyclic ligands as suitable neutral ionophores in selective complexation as well as in construction of selective electrodes for heavy metal ions including UO_2^{2+} *etc.* [18]. In this work as a preliminary study in designing of a biosensor, we report on the synthesis and antimicrobial evaluation of some macrocyclic crown ethers including amide, sulfonamide, and disulfide moieties (Scheme 1). We decided to synthesize the macrocyclic *bis*-sulfonamide. The *p*-chlorophenol which was employed as precursors in their preparation is an antiseptic drug. *p*-Chlorosulfonamide (chloroquine) is a potent antimalarial drug. Salicylanilides (2-hydroxy-*N*-phenylbenzamides) have been reported as a class of compounds with a wide variety of interesting biological activities, including antimycobacterial and antifungal effects [19-21].

2. Experimental

2.1. Chemicals and reagents

Chemicals were either prepared in our laboratories or purchased from Merck, Fluka and Aldrich Chemical Companies. All yields refer to isolated products. The reactions were monitored by thin-layer chromatography carried out on silica

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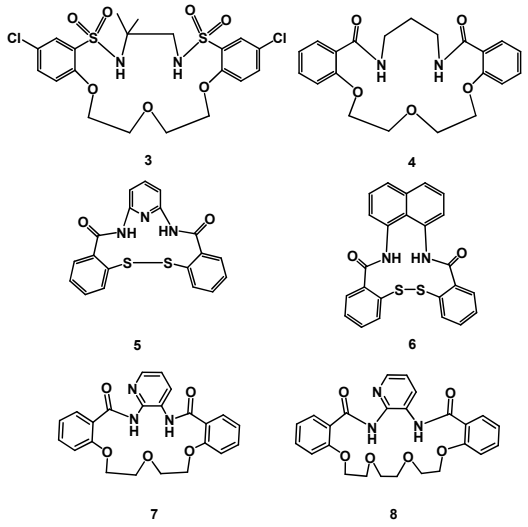
plates. The products were characterized by comparison of their physical data with those of known samples or by their spectral data. IR spectra were recorded on a Shimadzu-IR 470 spectrophotometer. ^{1}H NMR spectra was recorded on a Bruker-100 MHz spectrometer in $CDCl_3$ as the solvent and TMS as internal standard. Mass Spectra were determined on a Shimadzu GCMS-QP 1000 EX instrument at 70 eV. *Bis*-chlorides (**1a-d**) and macrocyclic bis-amides **3**, **5**, and **6** were prepared according to previously reported procedures [22-24]. The microorganisms (*Bacillus cereus* (PTCC1247), *Staphylococcus aureus* (PTCC1431), *Escherichia coli* (HB101BA7601C), *Pseudomonas aeruginosa* (PTCC1074), *Aspergillus niger* (PTCC5011) and *Fusarium oxysporum* (PTCC5115)) were purchased from Pasteur Institute of Iran. *Flammulina velutipes* was gifted from microbiology laboratory of Ferdowsi University of Mashhad.



Scheme 1

2.2. General procedure for the synthesis of macrocyclic bis-amides (3-8) by fast addition method

A solution of diamine **2a-e** (2.0 mmol) and triethylamine (0.41 g, 4.0 mmol) in $CHCl_3$ (10 mL) was added quickly (5 sec.) to a vigorously stirring solution of dichloride **1a-d** (2.0 mmol) in $CHCl_3$ (10 mL) at 0 °C. The reaction mixture was stirred at room temperature for 20 mins. The precipitate was filtered off and the filtrate washed with water (2 x 20 mL) and 10% aqueous sodium hydroxide solution (20 mL) and then with water (50 mL). The organic layer was dried with anhydrous magnesium sulfate and the solvent was evaporated to give a solid product. The crude product was purified by either recrystallization from petroleum ether and *n*-hexane or column chromatography using petroleum ether (B.p.: 60-80 °C)-ethyl acetate as eluent.

6,7,9,10,17,18,19,20,21,22-Decahydrodibenzo[h,r][1,4,7,11,15]trioxadiazacyclo-nonadecine-16,23-dione (4) was obtained from 1,3-diaminopropane **2b** (0.148 g, 2 mmol) and diacid chloride **1b** (0.76 g, 2 mmol). White. Yield: 95%. M.p.: 170-171 °C. FT-IR (KBr, cm^{-1}): 3300 ν(NH) (brs, amide), 1680 ν(C=O) (amide). ^{1}H NMR ($CDCl_3$, 100 MHz): 8.30 (b, 2H, NH), 8.20 (d, *J*=8.0 Hz, 2H, Ar-*H*), 7.40 (m, 2H, Ar-*H*), 6.9-7.2 (m, 4H, Ar-*H*), 4.2-4.5 (complex, 4H, Ar-*OCH$_2$*), 3.9-4.1 (complex, 4H, Ar-*OCH$_2$CH$_2$O*), 3.54 (complex, 4H, HN*CH$_2$*), 2.21 (m, 2H, HNCH$_2$*CH$_2$*). MS (EI, m/z(%)): 384 (M^+). Anal. Calcd. for $C_{21}H_{24}N_2O_5$: C, 65.61; H, 6.29; N, 7.29. Found: C, 65.53; H, 6.23; N, 7.19.

13,14,16,17-Tetrahydrodibenzo[h,q]pyrido[2,3-l][1,4,7,11,14]trioxadiazacyclooctadecine-7,23-dione (7) was obtained from 2,3-diaminopyridine **2e** (0.218 g, 2 mmol) and diacid chloride **1b** (0.76 g, 2 mmol). Pale yellow. Yield: 70%. M.p.: 138-139 °C; FT-IR (KBr, cm^{-1}): 3300 ν(NH) (brs, amide), 1675 ν(C=O) (amide). ^{1}H NMR ($CDCl_3$, 100 MHz): 9.75 (b, 1H, NH), 9.50 (b, 1H, NH), 8.25 (d, *J*=8.0 Hz, 1H, Ar-*H*), 8.10 (d, *J*=8.0 Hz, 1H, Ar-*H*), 6.8-7.6 (complex, 9H, Ar-*H*), 4.4 (complex, 2H, Ar-*OCH$_2$*), 4.1 (complex, 2H, Ar-*OCH$_2$*), 3.8 (complex, 4H, Ar-*OCH$_2$CH$_2$O*). MS (EI, m/z(%)): 419 (M^+). Anal. Calcd. for $C_{23}H_{21}N_3O_5$: C, 65.86; H, 5.05; N, 10.02. Found: C, 65.73; H, 5.03; N, 10.09.

6,7,9,10,12,13,25,26-Octahydrodibenzo[k,t]pyrido[2,3-o][1,4,7,10,14,17]tetraoxadiazacycloheni cosine-19,27-dione (8) was obtained from 2,3-diaminopyridine **2e** (0.218 g, 2 mmol) and diacid chloride **1d** (0.85 g, 2 mmol). Pale yellow. Yield: 81%. M.p.: 115-116 °C; FT-IR (KBr, cm^{-1}): 3340 ν(NH) (brs, amide), 1665 ν(C=O) (amide). ^{1}H NMR ($CDCl_3$, 100 MHz): 10.3 (b, 1H, NH), 9.9 (b, 1H, NH), 8.2 (dd, J_1=8.0 Hz, J_2=1.5 Hz, 1H, Ar-*H*), 8.05 (dd, J_1=8.0 Hz, J_2=1.5 Hz, 1H, Ar-*H*), 6.85-7.5 (complex, 9H, Ar-*H*), 4.4 (complex, 4H, Ar-*OCH$_2$*), 3.7 (complex, 4H, Ar-*OCH$_2$CH$_2$O*), 3.5 (complex, 4H, $CH_2OCH_2CH_2OCH_2$). MS (EI, m/z(%)): 463 (M^+). Anal. Calcd. for $C_{25}H_{25}N_3O_6$: C, 64.79; H, 5.44; N, 9.07. Found: C, 64.63; H, 5.23; N, 9.19.

2.3. Antimicrobial Screening

Reperesentative compounds **3**-**6** were screened *in vitro* for their antibacterial activities against bacteria (*Bacillus cereus* (PTCC1247), *Staphylococcus aureus* (PTCC1431), *Escherichia coli* (HB101BA7601C), and *Pseudomonas aeruginosa* (PTCC1074)) using disc diffusion method. Dimethyl sulfoxide (DMSO) was used, as a solvent to prepare desired solution (5%) of the selected compounds initially. The inhibition zones (mm) of microbial growth produced by different compounds (100 µg/disc) were measured (as a mean of three replicates) at the end of an incubation period of 24 h at 35 °C. *Streptomycin* was used as a standard antibiotic for the evaluation of the antimicrobial activity.

These compounds were screened for their antifungal activities against three fungal species (*Aspergillus niger* (PTCC5011), *Fusarium oxysporum* (PTCC5115)), and *Flammulina velutipes*). In the case of antifungal screening the concentration of tested compound in DMSO is also 5% and the inhibition zone (that obtained in each case as a mean of three replicates) were compared with fluconazole as a standard antifungal drug at the end of an incubation period of 72 h at 25°C.

3. Results and discussion

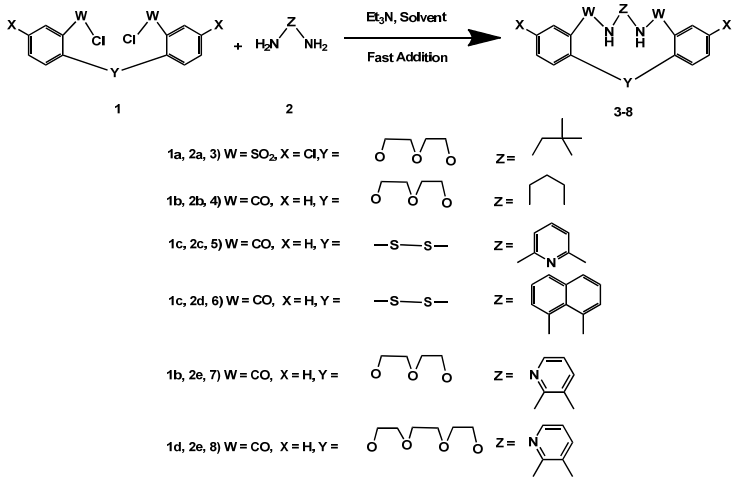
Typical macrocyclic bis-sulfonamide (**3**), amide (**4**, **7**, and **8**), and disulfides (**5** and **6**) (Scheme 1), are prepared by reacting *bis*-chlorides (**1a-d**) and diamines (**2a-e**) in a fast addition method [25,26] (Scheme 2). The cyclization was carried out with vigorously stirring and fast addition of a mixture of the diamine (**2a-e**) and triethylamine into a solution of *bis*-chlorides (**1a-d**) in $CHCl_3$ over 5 sec. A typical preparation of macrocyclic diamide (**4**) is as follow: a solution of the 1,3-diaminopropane (**2b**) (2 mmol) and triethylamine (4 mmol) in $CHCl_3$ (10 mL) was added rapidly (5 sec.) into a vigorously stirred suspension of diacid chloride (**1b**) (2 mmol) in $CHCl_3$ (10 mL) at 0 °C. After the usual work up and purification, dilactam macrocycle **4** was obtained in 95% yield. Macrocyclic crown ethers **3**, and **5**-**8** were obtained in a similar manner in good yields. The structures proposed for the macrocyclic compounds are consistent with data derived from IR and ^{1}H-NMR spectra and molecular weights that determined by mass spectrometric analysis.

In order to investigate the potential anti-microbial activities of macrocyclic compounds with sulfonamide and disulfide moieties, compounds **3**-**6**, as examples, were selected and screened *in vitro* for their antimicrobial activity against

Table 1. Response of various microorganisms to some synthesized macrocyclic crown ethers *in vitro*.

Compound	Bacteria Inhibition Zone (mm)				Fungi Inhibition Zone[a]		
	Bacillus cereus	*Staphylococcus aureus*	*Escherichia coli*	*Pseudomonas aeruginosa*	*Aspergillus niger*	*Flammulina velutipes*	*Fusarium oxysporum*
3	16	14	13	13	+	+	+
4	-	-	10	12	-	±	+
5	10	10	-	10	-	±	-
6	13	14	20	10	-	+	±
Streptomycin	22	13	17	14			
Fluconazole					++	+	+

[a] High activity (+); Moderate activity (±); Not activity (-).



Scheme 2

four strains of bacteria (*Bacillus cereus*, *Staphylococcus aureus, Escherichia coli* and *Pseudomonas aeruginosa*) and three fungal species (*Aspergillus niger, Flammulina velutipes* and *Fusarium oxysporum*). The antibacterial activities of tested compounds were evaluated by the reported method [27] using 5% concentration of selected compounds in DMSO as a solvent. The inhibition zones (mm) were compared with *Streptomycin* as a reference drug. In the case of antifungal the concentration of tested compound is also 5% and the inhibition zone were compared with fluconazole as a standard antifungal drug. The biological activities of the tested microorganisms are summarized in Table 1. From Table 1, it is obvious that, tested compounds show remarkable antibacterial activities while the antifungal activities are not remarkable except in the case of bis-sulfonamide **3** which reveals acceptable activity.

Compounds **3**, **5** and **6** show antibacterial activities against most strains tested. The results were compared with *Streptomycin* as a reference drug and dilactam **4** as a simple macrocycle without any sulfonamide or disulfide groups. Macrocyclic *bis*-sulfonamide **3** with reasonable antibiotic activity has two *m*-chlorosulfonamide moieties in its structure and also is similar to the *p*-chlorophenol, dichlorophenol, gamophene, chloroquine and Salicylanilides. Although the antibacterial activity involves multiple mechanisms [28], the antibacterial activity of these macrocycles may be attributed from one of the following issues;

a) The presence of acidic sulfonamide protons may change the pH, or react with basic moieties of proteins in the cell membrane and deformed the cells.

b) Complexation of macrocycles with metal ions in the cell and deformed the cell by uptake ions from them.

c) Complexation of macrocycles with metal ions in the cell membrane may inhibit the activity of those enzymes which activated in the presence of metal ions (diastases need a co-diastase such as Cu^{2+} ion).

d) Nucleophilic displacement of chloro group with NH_2 or SH residues of proteins in the cell membrane.

e) Disulfide (S-S) moiety of the macrocycles **5** and **6** may contributed in oxidative-reductive dehydrogenase reactions in the respiratory system and protoplast in the cell and so, waste the energy.

Since, macrocylic amide **4** does not show considerable activity, possibilities *b* and *c* are ruled out in the case of *bis*-sulfonamide **3**. On the other hand, sulfonamide protons are strong acids than amides. So, possibilities *a* and *d* are the suggested source of activity of compound **3**. While we suggest the moderate activities of disulfides **5** and **6** are due to the possibilities *a* or *e*.

4. Conclusion

In conclusion, sulfonamide and disulfide containing crown ethers revealed promising antibacterial activity. Besides sulfonamide crown ethers could be a good biosensor candidate for known microorganisms. Thus, these compounds could be used as lead structure in biosensor preparation and pharmaceutical industry.

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References

[1]. Blake, A. J.; Schröder, M. *Adv. Inorg. Chem.* **1990**, *35*, 1-80.
[2]. Balzani, V.; Credi, A.; Raymo, F. M.; Stoddart, F. *Angew. Chem. Int. Ed.* **2000**, *39*, 3349-3391.
[3]. Nabeshima, T.; Furusawa, H.; Tsukuda, N.; Shinnai, T.; Haruyama, T.; Yano, T. Y. *Heterocycles* **1995**, *41*, 655-659.

[4]. Cantor, C. R.; Schimell, P. R. Biophysical Chemistry, 1st Ed., Freeman, New York, 1980, Part I; pp. 293–295.
[5]. Ryser, H. J.; Levy, E. M.; Mandel, R.; DiSciullo, G. J. *Proc. Natl. Acad. Sci. USA* **1994**, *91*, 4559-4563.
[6]. Ranganathan, S.; Muraleedharan, K. M.; Bharadwaj, P.; Chatterji, D.; Karle, I. *Tetrahedron* **2002**, *58*, 2861-2874.
[7]. Nabeshima, T.; Nishida, D.; Saiki, T. *Tetrahedron* **2003**, *59*, 639-647.
[8]. Liu, Y.; Zhang, H. Y.; Chen, L. X.; He, X. W.; Wada, T.; Inoue, Y. *J. Org. Chem.* **2000**, *65*, 2870-2874.
[9]. Harusawa, S.; Yoshida, K.; Kojima, C.; Araki, L.; Kurihara, T. *Tetrahedron* **2004**, *60*, 11911-11922.
[10]. Sneader, W. Drug Discovery: A History, 1st Ed., John Wiley and Sons, 2005.
[11]. El-Atrouni, W. I.; Temesgen, Z. *Drugs Today* **2007**, *43*, 671-679.
[12]. Surleraux, D. L. N. G.; Tahri, A.; Verschueren, W. G.; Pille, G. M. E.; de Kock, H. A.; Jonckers, T. H. M.; Peeters, A.; De Meyer, S.; Azijn, H.; Pauwels, R.; de Bethune, M.; King, N. M.; Prabu-Jeyabalan, M.; Schiffer, C. A.; Wigerinck, P. B. T. P. *J. Med. Chem.* **2005**, *48*, 1813-1822.
[13]. Kostrowicki, J.; Luboch, E.; Makuch, B.; Cygan, A.; Horbaczewski, A.; Biernat, J. F. *J. Chromatog. A* **1988**, *454*, 340-344.
[14]. Bochenska, M.; Biernat, J. F.; Topolski, M.; Bradshaw, J. S.; Bruening, R. L.; Izatt, R. M. *J. Inclu. Phenom. Mol. Recog. Chem.* **1989**, *7*, 599-611.
[15]. Biernat, J. F.; Bradshaw, J. S.; Wilson, B. E.; Dalley, N. K.; Izatt, R. M. *J. Heterocyclic Chem.* **1986**, *23*, 1667-1671.
[16]. Bradshaw, J. S.; Koyama, H.; Dalley, N. K.; Izatt, R. M.; Biernat, J. F.; Bochenska, M. *J. Heterocyclic Chem.* **1987**, *24*, 1077-1083.
[17]. Biernat, J. F.; Bochenska, M.; Bradshaw, J. S.; Koyama, H.; Lindh, G.; Lamb, J. D.; Christensen, J. J.; Izatt, R. M. *J. Inclu. Phenom.* **1987**, *5*, 729-738.
[18]. Shamsipur, M.; Zargoosh, K.; Mizani, F.; Eshghi, H.; Rostami, F. *Spectrochimica Acta A* **2010**, *77*, 319-323.
[19]. Fluente, R. D. L.; Sonawane, N. D.; Arumainayagam, D.; Verkman, A. S. *Br. J. Pharmacol.* **2006**, *149*, 551-559.
[20]. Vinsova, J.; Imramovsky, A.; Buchta, V.; Ceckova, M.; Dolezal, M.; Staud, F.; Jampilek, J.; Kaustova, J. *Molecules* **2007**, *11*, 1-12.
[21]. Dahlgren, M. K.; Kauppi, A. M.; Olsson, I. M.; Linusson, A.; Elofsson, M. *J. Med. Chem.* **2007**, *50*, 6177-6188.
[22]. Eshghi, H. *Synth. Commun.* **2008**, *38*, 2540-2547.
[23]. Eshghi, H.; Seyedi, S. M.; Sandaroos, R. *Chin. Chem. Lett.* **2007**, *18*, 1439-1442.
[24]. Rahimizadeh, M.; Eshghi, H.; Rostami, F.; Faghihi, Z. *Polish J. Chem.* **2005**, *79*, 73-81.
[25]. Eshghi, H.; Mirzaie, M.; Esmaily-Shahry, H. *J. Chem. Res.* **2007**, 272-274.
[26]. Eshghi, H.; Bakavoli, M.; Hosseini, M. *J. Chem. Res.* **2006**, 740-743.
[27]. Mahfouz, N. M.; Moharram, A. M. *Pharm. Pharmacol. Commun.* **1999**, *5*, 315-322.
[28]. Macielag, M. J.; Demers, J. P.; Fraga-Spano, S. A.; Hlasta, D. J.; Johnson, S. G.; Kanojia, R. M.; Russell, R. K.; Sui, Z.; Weidner-Wells, M. A.; Werblood, H.; Foleno, B. D.; Goldschmidt, R. M.; Loeloff, M. J.; Webb, G. C.; Barrett, J. F. *J. Med. Chem.* **1998**, *41*, 2939-2945.