European Journal of Chemistry 5 (4) (2014) 635-638

European Journal of Chemistry

Journal homepage: www.eurjchem.com

Synthesis and characterization of four new unsymmetrical potentially pentadentate Schiff base ligands and related Zn(II) and Cd(II) complexes

Ahmad Ali Dehghani-Firouzabadi * and Fahimeh Motevaseliyan

Department of Chemistry, Faculty of Science, Yazd University, 89195-741 Yazd, Iran

**Corresponding author at: Department of Chemistry, Faculty of Science, Yazd University, 89195-741 Yazd, Iran.*
Tel.: +98.351.8122664. Fax: +98.351.8210644. E-mail address: aadehghani@yazd.ac.ir (A.A. Dehghani-Firouzabadi).

ARTICLE INFORMATION

DOI: 10.5155/eurjchem.5.4.635-638.1131

Received: 06 August 2014
Received in revised form: 23 August 2014
Accepted: 23 August 2014
Online: 31 December 2014

KEYWORDS

Diamine
Thioether
N2O2S ligands
Pentadentate ligand
Unsymmetric ligand
Schiff base complexes

ABSTRACT

Two thioether diamines 2-(2-aminoethylthio)aniline and 2-(3-aminopropylthio)aniline have been prepared employing a new procedure by reaction of 2-aminothiophenol and *N*-(2-bromoethyl)phthalimide or *N*-(3-bromopropyl)phthalimide, respectively. Then two new potentially pentadentate (N_2O_2S) Schiff base ligands derived from direct condensation between two maintained diamines and 2-hydroxybenzaldehyde or 5-bromo-2-hydroxy benzaldehyde were synthesized. Also, Zn(II) and Cd(II) complexes of Schiff base ligands have been prepared. These compounds have been characterized by physico-chemical and spectroscopic methods.

1. Introduction

Transition metal complexes with nitrogen, oxygen and sulfur donors ligands have been prepared since the beginning of the development of coordination chemistry and there is continuing interest in these complexes [1-3]. It has attracted more attention in metal complexes of unsymmetrical Schiff bases ligands with nitrogen, oxygen and sulfur atoms in recent years due the fact that the ligands around central metal ions in natural systems are unsymmetrical [4,5]. The presence of both hard and soft donor atoms in the backbones of unsymmetrical Schiff bases ligands, they readily coordinate with a wide range of transition metal ions [1-8]. Most of these unsymmetrical Schiff bases ligands obtained by the condensation of different types of primary amines with various ketones and aldehydes [9,10] by metal-templated [11-13] or by direct syntheses [14,15].

The synthesis of transition metal complexes of unsymmetrical Schiff bases ligands with nitrogen, oxygen and sulfur atoms is an important area of study with implications in bioinorganic chemistry [4,5,16], catalysis [17] and medical chemistry [18].

In this study, we report the preparation of 2-(2-amino ethylthio)aniline and 3-(2-aminopropylthio)aniline in new procedure (Scheme 1) and four new macroacyclic pentadentate (N_2O_2S) unsymmetrical Schiff base ligands by direct condensation of the 2-(2-aminoethylthio)aniline or 3-(2-aminopropylthio)aniline and 2-hydroxybenzaldehyde or 5-bromo-2-hydroxybenzaldehyde (Scheme 2). Zn(II) and Cd(II) complexes of these unsymmetrical Schiff base ligands have been synthesized.

2. Experimental

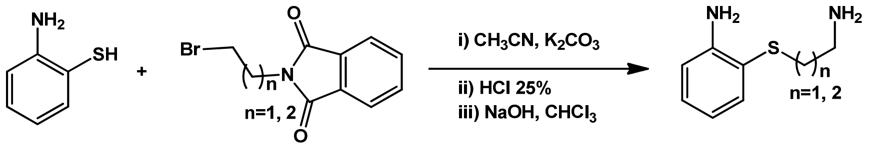
2.1. Chemical and starting materials

2-Aminothiophenol, 2-hydroxybenzaldehyde, 5-bromo-2-hydroxybenzaldehyde, $Zn(NO_3)_2\cdot 6H_2O$ and $Cd(NO_3)_2\cdot 4H_2O$ were purchased from Merck and used as received. Other reagents and solvents used were of analytical grade and purchased commercially.

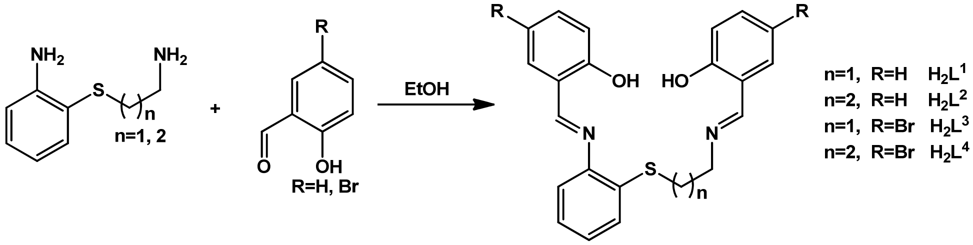
2.2. Instrumentation

Elemental analyses were performed in a CHNS-O-2400 II Perkin-Elmer. Infrared spectra were recorded in ATR, using a Bruker FT-IR Equinax-55 spectrophotometer (4000-400 cm^{-1}). Mass spectra were obtained using a QP-1100EX Shimadzu GC–MS (EI at 70 eV). ^{1}H and ^{13}C NMR spectra were taken in $CDCl_3$ on a Bruker NMR 500 MHz spectrometer using $Si(CH_3)_4$ as an internal standard.

ISSN 2153-2249 (Print) / ISSN 2153-2257 (Online)
http://dx.doi.org/10.5155/eurjchem.5.4.635-638.1131



Scheme 1



Scheme 2

2.3. General synthesis of the thioether diamines

2-Aminothiophenol (6.25 g, 50 mmol) was dissolved in acetonitrile (40 mL) and K_2CO_3 (6.9 g, 50 mmol) was added. The mixture was refluxed and then a solution of *N*-(2-bromo ethyl)phthalimide (12.65 g, 50 mmol) or *N*-(3-bromopropyl) phthalimide (13.4 g, 50 mmol) in acetonitrile (40 mL) was added. The mixture was refluxed for 8 h and then filtered hot. The filtrate was reduced to dryness by rotary evaporation. The resulting product was boiled under reflux for 8 h in aqueous HCl (25%, 100 mL). After this time the solution was evaporated to small volume (ca. 25 mL) under vacuum and cooled in refrigerator for several hours. The solid present was filtered off, and the filtrate was evaporated to dryness under vacuum. Water (50 mL) was added to the mixture and the pH was adjusted to 13 with sodium hydroxide. The product was extracted with chloroform (3 × 25 mL); the combined chloroform solutions were separated and dried over magnesium sulphate. The chloroform was removed by rotary evaporation to leave brown oil (Scheme 1).

2-(2-Aminoethylthio)aniline: Yield: 5.63 g (67%). FT-IR (ATR, ν, cm^{-1}): 3430, 3352, 3172 (NH_2), 750 (C-S). ^1H NMR (500 MHz, $CDCl_3$, δ, ppm): 2.81 (s, 4H, NCH_2CH_2S), 3.30-5.20 (br, 4H, NH_2), 6.68 (t, 1H, *J* = 7.3 Hz, Ar-*H*), 6.72 (d, 1H, *J* = 8.0 Hz, Ar-*H*), 7.12 (t, 1H, *J* = 6.6 Hz, Ar-*H*), 7.38 (d, 1H, *J* = 6.3 Hz, Ar-*H*). ^{13}C NMR (125 MHz, $CDCl_3$, δ, ppm): 38.8 (1C, CCH_2S), 41.4 (1C, NCH_2C), 115.4, 117.3, 118.9, 130.3, 136.6, 148.9 (6C, Ar-*C*).

2-(3-Aminopropylthio)aniline: Yield: 6.55 g (72%). FT-IR (ATR, ν, cm^{-1}): 3432, 3338, 3172 (NH_2), 746 (C-S). ^1H NMR (500 MHz, $CDCl_3$, δ, ppm): 1.73 (m, 2H, *J* = 7.2 Hz, CCH_2C), 2.81 (t, 2H, *J* = 7.3 Hz, NCH_2C), 2.84 (t, 2H, *J* = 5.8 Hz, CCH_2S), 3.50-5.00 (br, 4H, NH_2), 6.70 (t, 1H, *J* = 7.5 Hz, Ar-*H*), 6.73 (d, 1H, *J* = 8.0 Hz, Ar-*H*), 7.13 (t, 1H, *J* = 7.9 Hz, Ar-*H*), 7.38 (d, 1H, *J* = 6.3 Hz, Ar-*H*). ^{13}C NMR (125 MHz, $CDCl_3$, δ, ppm): 32.5 (1C, CCH_2C), 32.9 (1C, CCH_2S), 40.9 (1C, NCH_2C), 115.4, 118.1, 118.9, 130.1, 136.2, 148.7 (6C, Ar-*C*).

2.4. General synthesis of the Schiff base ligands

A solution of 2-(2-aminoethylthio)aniline or 2-(3-amino propylthio)aniline (5 mmol) in ethanol (50 mL) was added dropwise to a refluxing solution of 2-hydroxybenzaldehyde or 5-bromo-2-hydroxybenzaldehyde (10 mmol) in the same solvent (40 mL) and four new Schiff base ligands, **H_2L^1-H_2L^4** were obtained, respectively. After refluxing for 3 h the solution was vacuum evaporated to yield the crude product as yellow oil. A small volume of petroleum ether was added to the residue remaining in the flask and rubbed. Then liquid was decanted and residue was evaporated until a yellow oil remained (Scheme 2).

2-((2-(2-(2-Hydroxybenzylidenamino)ethylthio)phenylimino) methyl)phenol (**H_2L^1**): Yield: 1.56 g (83%). FT-IR (ATR, ν, cm^{-1}): 2500-3000 (OH) (br), 1629, 1612 (C=N), 750 (C-S). ^1H NMR (500 MHz, $CDCl_3$, δ, ppm): 3.27 (t, 2H, NCH_2), 3.86 (t, 2H, CH_2S), 6.8-7.5 (m, 12H, Ar-*H*), 8.33 (s, 1H, CH=N), 8.60 (s, 1H, CH=N), 13.09 (br, 1H, OH), 13.28 (s, 1H, OH). ^{13}C NMR (125 MHz, $CDCl_3$, δ, ppm): 34.0 (1C, CH_2S), 58.7 (1C, NCH_2); 117.1, 117.4, 117.9, 118.6, 118.7, 119.1, 119.3, 126.4, 127.6, 127.8, 131.5, 132.3, 132.5, 132.8, 133.3, 146.6, 160.2, 160.9, (18C, Ar-*C*), 162.8, 165.1 (2C, CH=N). EI-MS (*m/z*, %): 376 [M^+, 100].

2-((2-(3-(2-Hydroxybenzylidenamino)propylthio)phenyl imino)methyl)phenol (**H_2L^2**): Yield: 1.46 g (75%). FT-IR (ATR, ν, cm^{-1}): 2500-3000 (OH) (br), 1629, 1611 (C=N), 750 (C-S). ^1H NMR (500 MHz, $CDCl_3$, δ, ppm): 2.11 (m, 2H, CCH_2C), 3.03 (t, 2H, NCH_2), 3.75 (t, 2H, CH_2S), 6.80-7.40 (m, 12H, Ar-*H*), 8.36 (s, 1H, CH=N), 8.62 (s, 1H, CH=N), 13.29 (br, 1H, OH), 13.39 (s, 1H, OH). ^{13}C NMR (125 MHz, $CDCl_3$, δ, ppm): 29.7 (1C, CCH_2C), 29.8 (1C, CH_2S), 57.9 (1C, NCH_2), 117.0, 117.4, 117.8, 118.6, 118.8, 119.2, 119.3, 126.5, 127.5, 127.9, 131.4, 132.3, 132.4, 132.5, 133.4, 146.6, 161.2, 162.2 (18C, Ar-*C*), 163.0, 165.8 (2C, CH=N). EI-MS (*m/z*, %): 390 [M^+, 100].

4-Bromo-2-((2-(2-(5-bromo-2-hydroxybenzylidenamino) ethylthio)phenylimino)methyl)phenol (**H_2L^3**): Yield: 2.23 g (86%). FT-IR (ATR, ν, cm^{-1}): 2500-3000 (OH) (br), 1634, 1614 (C=N), 757 (C-S). ^1H NMR (500 MHz, $CDCl_3$, δ, ppm): 3.12 (t, 2H, NCH_2), 3.86 (t, 2H, CH_2S), 6.60-7.80 (m, 10H, Ar-*H*), 8.30 (s, 1H, CH=N), 8.63 (s, 1H, CH=N), 13.21 (br, 1H, O-H), 13.32 (s, 1H, O-H). ^{13}C NMR (125 MHz, $CDCl_3$, δ, ppm): 33.9 (1C, CH_2S), 58.5 (1C, NCH_2), 110.1, 110.7, 117.7, 119.1, 119.3, 120.1, 120.6, 126.6, 127.9, 128.2, 132.7, 133.6, 134.3, 134.7, 135.8, 146.3, 160.1, 160.3 (18C, Ar-*C*), 160.6, 164.7 (2C, CH=N). EI-MS (*m/z*, %): 534 [M^+, 100].

4-Bromo-2-((2-(3-(5-bromo-2-hydroxybenzylidenamino) propylthio)phenylimino)methyl)phenol (**H_2L^4**): Yield: 1.97 g (72%). FT-IR (ATR, ν, cm^{-1}): 2500-3000 (OH) (br), 1632, 1612 (C=N), 752 (C-S). ^1H NMR (500 MHz, $CDCl_3$, δ, ppm): 2.09 (m, 2H, CCH_2C), 3.00 (t, 2H, NCH_2), 3.74 (t, 2H, CH_2S), 6.8-7.7 (m, 10H, Ar-*H*), 8.26 (s, 1H, CH=N), 8,53 (s, 1H, CH=N), 13.31 (br, 1H, OH), 13.39 (s, 1H, OH). ^{13}C NMR (500 MHz, $CDCl_3$, δ, ppm): 29.4 (1C, CCH_2C), 29.9 (1C, CH_2S), 57.8 (1C, NCH_2), 110.0, 110.6, 117.8, 119.0, 119.4, 120.0, 120.7, 126.6, 128.0, 128.2, 132.6, 133.5, 134.3, 134.9, 135.9, 146.1, 160.1, 160.2, (18C, Ar-*C*), 160.4, 164.6 (2C, CH=N). EI-MS (*m/z*, %): 548 [M^+, 100].

2.5. General synthesis of the complexes

A solution of $Zn(NO_3)_2\cdot6H_2O$ or $Cd(NO_3)_2\cdot4H_2O$ (1 mmol) and NEt_3 (2 mmol) in ethanol (30 mL) was added to a refluxing solution of H_2L^n (n = 1, 2, 3, 4) (1 mmol) in the same solvent (30 mL) and the reaction mixture was refluxed for 2 h. The reaction mixture was then concentrated to *ca.* 5-10 mL. The complexes were filtered off, washed with cold ethanol and air-dried.

[Zn(HL¹)]NO₃·EtOH: Yield: 252 mg (46%). FT-IR (ATR, ν, cm^{-1}): 1607 (C=N), 754 (C-S). ^1H NMR (500 MHz, $CDCl_3$, δ, ppm): 3.23 (t, 2H, NCH_2), 3.83 (t, 2H, CH_2S), 6.40-7.70 (m, 12H, Ar-*H*), 8.10 (br, 2H, CH=N), 10.90 (br, 1H, OH). EI-MS (*m/z*, %): 439 $[Zn(HL^1)]^+$. Anal. calcd. for $C_{24}H_{25}N_3O_6SZn$: C, 52.51; H, 4.59; N, 7.65. Found: C, 51.78; H, 4.30; N, 7.77%.

[Zn(HL²)]NO₃·EtOH: Yield: 293 mg (52%). FT-IR (ATR, ν, cm^{-1}):) 1634 (C=N), 746 (C-S). EI-MS (*m/z*, %): 454 $[Zn(HL^2)]^+$. Anal. calcd. for $C_{25}H_{27}N_3O_6SZn$: C, 53.34; H, 4.83; N, 7.46. Found: C, 53.57; H, 5.13; N, 7.69%.

[Zn(HL³)]NO₃·EtOH: Yield: 465 mg (66%). FT-IR (ATR, ν, cm^{-1}): 1605 ν(C=N), 754 ν(C-S). EI-MS (*m/z*, %): 598 $[Zn(HL^3)]^+$. Anal. calcd. for $C_{24}H_{23}Br_2N_3O_6SZn$: C, 40.79; H, 3.28; N, 5.95%. Found: C, 41.24; H, 3.57; N, 5.78%.

[Zn(HL⁴)]NO₃·EtOH: Yield: 497 mg (69%). FT-IR (ATR, ν, cm^{-1}): 1634, 1611 (C=N), 746 (C-S). EI-MS (*m/z*, %): 611 $[Zn(HL^4)]^+$. Anal. calcd. for $C_{25}H_{25}Br_2N_3O_6SZn$: C, 41.66; H, 3.50; N, 5.83. Found: C, 41.41; H, 3.44; N, 6.04%.

[Cd(HL¹)]NO₃: Yield: 435 mg (79%). FT-IR (ATR, ν, cm^{-1}): 1607 (C=N), 755 (C-S). EI-MS (*m/z*, %): 487 $[Cd(HL^1)]^+$. Anal. calcd. for $C_{22}H_{19}N_3O_5SCd$: C, 48.05; H, 3.48; N, 7.64. Found: C, 47.59; H, 3.42; N, 7.29%.

[Cd(HL²)]NO₃: Yield: 407 mg (72%). FT-IR (ATR, ν, cm^{-1}): 1622 (C=N), 752 (C-S). ^1H NMR (500 MHz, $CDCl_3$, δ, ppm): 2.10 (m, 2H, CCH_2C), 3.01 (t, 2H, NCH_2), 3.79 (t, 2H, CH_2S), 6.40-7.70 (m, 12H, Ar-*H*), 8.10 (br, H, CH=N), 8.30 (br, 1H, CH=N), 11.70 (br, 1H, O-H). EI-MS (*m/z*, %): 503 $[Cd(HL^2)]^+$. Anal. calcd. for $C_{23}H_{21}N_3O_5SCd$: C, 48.99; H, 3.75; N, 7.45. Found: C, 49.45; H, 3.72; N, 6.93%.

[Cd(HL³)]NO₃: Yield: 601 mg (85%). FT-IR (ATR, ν, cm^{-1}): 1610, 1620 (C=N), 759 (C-S). EI-MS (*m/z*, %): 644 $[Cd(HL^3)]^+$. Anal. calcd. for $C_{22}H_{17}Br_2N_3O_5SCd$: C, 37.34; H, 2.42; N, 5.94. Found: C, 36.90; H, 2.19; N, 6.44%.

[Cd(HL⁴)]NO₃: Yield: 560 mg (86%). FT-IR (ATR, ν, cm^{-1}): 1627, 1610 (C=N), 747 (C-S). EI-MS (*m/z*, %): 660 $[Cd(HL^4)]^+$. Anal. calcd. for $C_{23}H_{19}Br_2N_3O_5SCd$: C, 38.28; H, 2.65; N, 5.82. Found: C, 37.99; H, 2.69; N, 5.26%.

3. Results and discussion

The IR spectrum for the 2-(2-aminoethylthio)aniline shows bands at 3430, 3352, 3172 cm^{-1} and for the 2-(3-aminopropyl thio)aniline shows bands at 3432, 3338, 3172 cm^{-1} assignable to the aromatic and aliphatic primary amine stretch. The three branches peaks in the area of 3150-3450 cm^{-1} (due to the interaction of the two groups together, three peaks is observed) show two different groups of NH_2 in the compounds. In addition, the vibration peak of C-S is observed in 750 and 746 cm^{-1}, respectively. The IR spectra for the ligands (H_2L^n, n = 1, 2, 3, 4) confirm the presence of imines (*ca.* 1611-1634 cm^{-1}) and the absence of carbonyl and amine functional groups of the starting materials. The total absence of ν(C=O) absorption in the IR spectra of the ligands together with the appearance of new ν(C=N) absorption in the range of 1611-1634 cm^{-1} clearly indicated that a new Schiff base ligand had formed in each case. In addition, the vibration peaks of C-S are observed in 750-757 cm^{-1}. The broad band observed in the spectra of ligands in the region of 2500-3000 cm^{-1} can be assigned to the ν(O-H) groups. The IR spectra of Zn(II) and Cd(II) complexes of these unsymmetrical Schiff base ligands show one or two strong band at 1605-1634 cm^{-1} assigned to the C=N stretching mode. These bands are shifted relative to the similar bands of the ligands that show the imines nitrogen atoms coordinated to the metal ions. The vibration peaks of C-S in complexes are observed in 746-759 cm^{-1}.

^1H NMR and $^{13}C\{1H\}$ NMR results, obtained for some prepared compounds at ambient temperature in $CDCl_3$, are presented in experimental section. The aliphatic protons appear as a singlet at 2.81 ppm in 2-(2-aminoethylthio)aniline and in related ligands (**H_2L^1** and **H_2L^3**) appear as two triplet resonance in the region δ 3.12-3.86 ppm but for 2-(3-aminopropylthio)aniline and related ligands (**H_2L^2** and **H_2L^4**) appear as three signals in the region δ 1.73-3.75 ppm. The broad signal at δ 3.3-5.2 ppm in the ^1H NMR spectra of diamines are assigned to the NH_2 protons and at δ 13.09-13.39 ppm in the ^1H NMR spectra of ligands are assigned to the OH protons. Signals for aromatic protons observed at δ 6.6-7.8 ppm. The ^1H NMR spectra of ligands, show peaks at δ 8.26-8.63 ppm corresponding to the imine protons and these indicates that the condensation has occurred. The ^1H NMR spectra of Zn(II) and Cd(II) complexes showed that the imine signals shifted in compared to initial compound. Unfortunately, the poor solubility of the complexes made impossible there's study by NMR spectroscopy.

The EI mass spectra of the unsymmetrical Schiff bases ligands, provide strong evidence for the formation of these ligands. The peaks in the spectra of the ligands are observed at *m/z* 376, 390, 534, 548 corresponding to $[H_2L^n]^+$ (n = 1, 2, 3, 4), respectively. The spectra for the complexes exhibit peaks at higher molecular weights, confirming the presence of metal unit in the complexes.

4. Conclusion

We report the successful synthesis of two thioether diamines 2-(2-aminoethylthio)aniline and 2-(3-aminopropyl thio)aniline and also four potentially pentadentate (N_2O_2S) unsymmetrical Schiff bases ligands (H_2L^n, n = 1, 2, 3, 4) have been prepared by reaction between of these diamines and 2-hydroxybenzaldehyde or 5-bromo-2-hydroxybenzaldehyde. The complexation capacity of four ligands towards Zn(II) and Cd(II) ions has been also investigated. The complexes are mononuclear and the spectroscopic data confirm the formation of them.

Acknowledgements

We are grateful to the Faculty of Chemistry of Yazd University and Ministry of Science, Research and Technology of Iran, for financial support.

References

[1]. Lindoy, L. F. *Coord. Chem. Rev.* **1969**, *4*, 41-71.
[2]. Sudbrake, C.; Vahrenkamp, H. *Eur. J. Inorg. Chem.* **2001**, 751-754.
[3]. Masdeu-Bulto, A. M.; Dieguez, M.; Martin, E.; Gomez, M. *Coord. Chem. Rev.* **2003**, *242*, 159-201.
[4]. Taki, M.; Hattori, H.; Osako, T.; Nagatomo, S.; Shiro, M.; Kitagawa, T.; Itoh, S. *Inorg. Chim. Acta* **2004**, *357*, 3369-3381.
[5]. Dolega, A. *Coord. Chem. Rev.* **2010**, *254*, 916-937.
[6]. Canovese, L.; Chessa, G.; Visentin, F.; Uguagliati, P. *Coord. Chem. Rev.* **2004**, *248*, 945-954.
[7]. Asadi, M.; Mohammadi, K.; Esmaiezadeh, S.; Etemadi, B.; Fun, H. K. *Inorg. Chim. Acta* **2009**, *362*, 4913-4920.
[8]. Chattopadhyay, S.; Ray, M. S.; Chaudhuri, S.; Mukhopadhyay, G.; Bocelli, G.; Cantoni, A.; Ghosh, A. *Inorg. Chim. Acta* **2006**, *359*, 1367-1375.
[9]. Matsushita, T.; Shono, T. *Polyhedron* **1986**, *5*, 735-743.
[10]. Rezaeivala, M.; Keypour, H.; Salehzadeh, S.; Latifi, R.; Chalabian, F.; Katouzian, F. *J. Iran Chem. Soc.* **2014**, *11*, 431-440.
[11]. Kaasjager, V. E.; Puglisi, L.; Bouwman, E.; Driessen, W. L.; Reedijk, J. *Inorg. Chim. Acta* **2000**, *310*, 183-190.
[12]. Keypour, H.; Dehghani-Firouzabadi, A. A.; Khavasi, H. R. *Polyhedron* **2009**, *28*, 1546-1550.
[13]. Keypour, H.; Dehghani-Firouzabadi, A. A.; Rezaeivala, M.; Goudarziafshar, H. *J. Iran Chem. Soc.* **2010**, *7*, 820-824.
[14]. Chiswell, B.; Crawford, J. P.; O'Reilly, E. J. *Inorg. Chim. Acta* **1980**, *40*, 223-228.

[15]. Costes, J. P. *Inorg. Chim. Acta* **1987**, *130*, 17-21.
[16]. Harrop, T. C.; Mascharak, P. K. *Acc. Chem. Res.* **2004**, *37*, 253-260.
[17]. Kwong, H. L.; Yeung, H. L.; Yeung, C. T.; Lee, W. S.; Lee, C. S.; Wong, W. L. *Coord. Chem. Rev.* **2007**, *251*, 2188-2222.
[18]. Saghatforoush, L. A, ; Chalabian, F.; Aminkhani, A.; Karimnezhad, G.; Ershad, S. *Eur. J. Med. Chem.* **2009**, *44*, 4490-4495.