The effect of Mn^{2+}, Ni^{2+} and Cu^{2+} Ions on Antibiotic Activity of Cephradine; Theoretical Study

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Abstract

Natural bond orbital (NBO) calculation based on the hybrid functional b3lyp/6-31G* (d) were performed to investigate the effect of Mn^{2+}, Ni^{2+} and Cu^{2+} ions on the monocyclic β-lactams; cephradine (Ceph) activity. The results show that, only Cu2+-cephradine complex has more antibacterial activity than the free Ceph. In contrast, monocationic bidentate Ceph-metal complexes with Mn^{2+}, Ni^{2+} ions have lower bacterial toxicity than the free Ceph.

Keywords: Ab initio calculation; β-lactam antibiotics; Cephradine metal complexes.

INTRODUCTION

Among antibiotic drugs, β-lactam antibiotics are well known and different kinds of them are utilized for treatment of a wide variety of bacterial infections [1-6]. The β-lactam antibiotics that are currently available all have a β-lactam ring system, a highly strained and reactive cyclic amide. The five relevant ring systems which appear in active β-lactam drugs are included the penam, penem, cephem and monobactam ring structures (scheme1).

Due to the elegant research it has been well established that β-lactam drugs have deathly effects on bacteria by inhibiting transpeptidation process required to complete the synthesis of peptidoglycan component of the bacterial cell walls. Biosynthesis of bacterial cell walls is an enzyme reaction catalyzed by high molecular weight penicillin-binding proteins (PBPs). In the final step of peptidoglycan synthesis, the serine hydroxyl of PBPs attacks the amide linkage of the D-alaD-ala segments of the amino acid side chain of peptidoglycan strands (scheme2). This process forms enzyme bonded intermediate that is finally displaced by the glycine amino terminus of a second strand. The highly strained and reactive β-lactam ring is able to react with the serine hydroxyl group of PBPs. The special arrangement of the β-lactam ring system closely resembles the conformation of the D-ala-D-ala segments of the peptidoglycan strand

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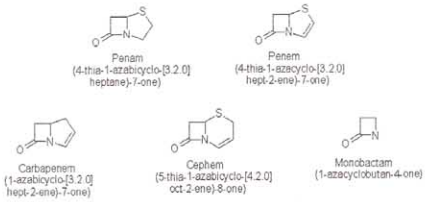
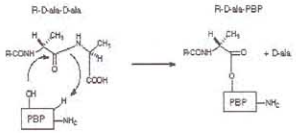


Scheme1. The most common ring systems appeared in β-lactam drugs

and the PBPs recognize it as a natural substrate. Reaction of the peptidoglycan with β-lactam thus results in an irreversible inhibition of the PBP and results in lysis and death of the bacterial cell.

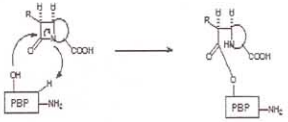


Scheme 2: Biosynthesis of bacterial cell walls mechanism catalyzed by PBPs

One of the most common species of β-lactam antibiotics are cephalosporins. The naturally occurring cephalosporins are 7-amino-8-oxo-5-thia-7-aza bicycle [4.2.0]oct-2-ene-2-carboxylic acids. The cephalosporines are similar to the penicillins in many aspects including several structural features and mode of action against bacteria. Most early cephalosporines being active against some gram negative bacteria as well as gram positive organisms [7,8]. The structure of cephradine and PBP deactivation mechanism by Ceph has been shown in scheme 3.

On the basis of biological investigations, pharmaceutical property of the many drugs generally modified when they used in the form of metal complexes [9-12]. The most widely studied metal complex in this respect is copper (II), which has proved beneficial in diseases such as tuberculosis, gastric ulcers, rheumatoid arthritis and cancers [13–16]. These results encouraged us to investigate the chemistry of metal-antibiotics complexes, e.g. $[M(Ceph)]^{n+}$ complexes with different transition metal ions.

In this theoretical work, we attempted to study the effect of Mn^{2+}, Ni^{2+} and Cu^{2+} ions on the structural characteristics of Ceph in relation to antibiotic activity.



Scheme 3. Deactivation mechanism of PBP by Ceph

Methods of calculation

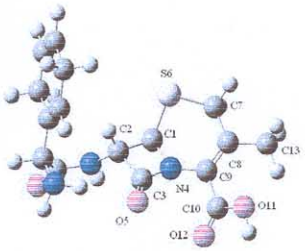
Geometry optimizations of Ceph and their related metal ion complexes were carried out without any symmetry limitation using B3LYP method, all with 6-31G*(d) basis set. The resulting geometries, obtained from this calculation were then verified as minima by frequency calculation and used to perform natural bond orbital (NBO) analysis by NBO 3.1 program as implemented in Gaussian03 using B3LYP method.

Results and discussion

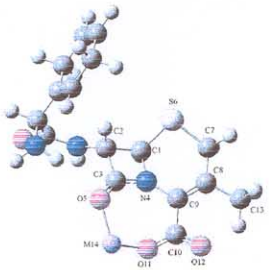
Optimized geometries of Ceph and their related metal complexes structural parameters at the B3LYP level of theory using 6-31G*(d) basis set, is shown in scheme 4, Table 1 and 2 respectively.

The results in Table 1 show that, Ceph is coordinated to metal ion through the carboxylic and lactamic oxygen in its anionic form. The lactamic carbonyl bond length (C_3-O_5) in the β-lactam ring-metal system, due to the metal coordination, increases from 1.21A^0 in free Ceph to 1.27A^0, 1.26A^0 in [Mn-Ceph]$^+$ and [Ni-Ceph]$^+$ and 1.22A^0 in [Cu-Ceph]$^+$.

The results reported in Table 1 show also that, C_3-N_4 bond length in Ceph-Cu due to the metal coordination, becomes slightly longer than free Ceph (viz.



(a)

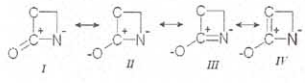


(b)

Scheme 4: Structure of (a) Ceph, (b) Ceph-metal complexes (M_{14}= Mn^{2+}, Ni^{2+}, Cu^{2+}).

1.40A versus 1.39) but, much more shorter in [Mn-Ceph]+ and [Ceph-Ni]+ (1.34 and 1.33 versus 1.39A). These results, in consistence with bond orders results (Table 3) can be ascribed to the formation of form III in [Mn-Ceph]$^+$ and [Ni-Ceph]$^+$ and form II in [Cu-Ceph]$^+$

complex (Scheme 5) due to the transition metal coordination. Formation of the resonance forms, I-IV have already proposed [17]. Whereas formation of form II and III due to metal coordination can be confirmed and verified by referring to the calculated β-lactam ring, C_3-N_4 bond orders, shown in Table 3. Accordingly, the C_3-N_4 bond order, increases from 1.14 in free Ceph (form I) to 1.33, 1.33 in [Mn-Ceph]+, [Ni-Ceph]+ (form III), and 1.04 in [Cu-Ceph]+(form II). According to these results we can ascribe that, weakening of



Scheme 5.

The C_3-N_4 bond in [Cu-Ceph]+ (form II), due to metal coordination of Ceph, in contrast to [Mn-Ceph]+ and [Ni-Ceph], facilitates the C_3-N_4 bond cleavage of β-lactam ring and enhances the antibiotic activity of Ceph referring to the hydrolysis of β–lactam ring by PBPs and the proposed mechanism of reaction that during the hydrolysis of Ceph, serine hydroxyl group of PBP attacks the β-lactam's carbonyl carbon and forms a tetrahedral intermediate followed by the C_3-N_4 bond cleavage of β-lactam ring with a concurrent intra molecular transfer of the hydroxide proton to the N_4 atom. So, we can conclude that, Cu2+-Ceph complex has more antibacterial activity than the free Ceph. This result is in accord with experimental data [12,16].

Table 4 shows the natural population analysis (NPA) evaluated in terms of natural orbital occupancies that reveals the percentage contribution of the molecular charge distribution. This Table also shows that, the electric positive atomic charge value on β-lactam carbonyl carbon atom increases from 0.71 for free Ceph to 0.724, 0.725 and 0.75 in [Mn-Ceph]+, [Ni-Ceph]+, and [Cu-Ceph]+ complexes respectively. This increase of positive charge density on β-lactam carbonyl carbon makes all metal-Ceph complexes, more suitable toward nucleophilic attack by alkaline hydrolysis and reflects enhancement of the antibacterial reactivity of Ceph due to metal coordination.

Table 5 shows the resulted natural atomic hybrids (NHO), on all atoms of β-lactam ring in metal-Ceph complexes with the polarization coefficient (in parentheses), for each hybrid in the corresponding NBO. The results reveal the difference in bonding capability between carbon and nitrogen atoms in β-lactam ring under strained conditions according to the different metal ion coordination. We also find out that, the C_3-N_4 bond in $[Cu\text{-}Ceph]^{2+}$ has weaker bond energy than that of Ceph, $[MnCeph]^{+}$ and $[Ni\text{-}Ceph]^{+}$ respectively (-0.801, -0.814 -1.014 and -1.034 Kcal/mol). We can attribute the difference between these values to the rehybridization effect from the strain [18]. A higher strain energy can be satisfactorily, attributed to the weaker rehybridization capability of carbon and nitrogen atoms in $[Cu\text{-}Ceph]^{2+}$.

Conclusion

On the basis of obtained results we conclude that, carbonyl group of β-lactam ring in Cu-Ceph ion complex is a more suitable site for PBPs nucleophilic attack and its –CO-N bond cleft easier than free Ceph. So β-lactam ring hydrolysis accomplishes easier and its

ablity to inhibit PBPs activity is better than free Ceph. In contrast, monocationic bidentate Ceph-metal complexes with Mn2+, Ni2+ ions have lower bacterial toxicity than the free Ceph.

From the NBO analysis we find out that, the s and p character of C_3-N_4 bond to be useful for examining rehybridization effect on the strain energy. The results show also the difference in bonding capability between carbon and nitrogen due to the different metal ion coordination, under strained conditions. We contributed the higher strain energy in the [Ceph-Cu]+ ion complex to a weaker rehybridization capability of nitrogen relative to carbon and easier –CO-N bond cleavage or thereby its high antibacterial activity in comparison to Ceph, $[MnCeph]^+$, $[Ni\text{-}Ceph]^+$ was concluded. Whereas the monocationic bidentate Ceph-metal complexes with Mn^{2+}, Ni^{2+} ions will have a lower bacterial toxicity than the free Ceph.

Table1. Selected bond lengths (Å) of Ceph and its metal complexes calculated using B3LYP level of theory and 6-31G (d, p) basis set

	Ceph	$[Ceph\text{-}Mn]^+$	$[Ceph\text{-}Ni]^+$	$[Ceph\text{-}Cu]^+$
C1-C2	1.56	1.55	1.55	1.54
C2-C3	1.55	1.52	1.52	1.52
C1-N4	1.46	1.49	1.49	1.49
C1-S6	1.83	1.804	1.804	1.81
C3-N4	1.39	1.34	1.33	1.40
N4-C9	1.40	1.43	1.42	1.38
C3-O5	1.21	1.27	1.26	1.22
S6-C7	1.84	1.84	1.84	1.83
C7-C8	1.52	1.52	1.52	1.51
C8-C9	1.35	1.36	1.36	1.39
C8-C13	1.51	1.50	1.50	1.49
C9-C10	1.50	1.501	1.48	1.53
C10-O11	1.35	1.35	1.35	1.26
C10-O12	1.21	1.21	1.21	1.24
O5-M14	-	1.95	1.80	1.99
O11-M14	-	1.83	1.74	1.93

Table2: Selected bond and dihedral angles in Ceph and its metal complexes calculated using B3LYP level of theory and 6-31G (d, p) basis set

	Ceph	$[Ceph-Mn]^+$	$[Ceph-Ni]^+$	$[Ceph-Cu]^+$
C2-C1-N4	88.62	87.82	87.51	88.35
C2-C1-S6	116.722	115.61	115.22	113.89
C1-C2-C3	85.132	84.32	84.09	86.23
C2-C3-N4	91.11	94.83	95.03	92.60
C2-C3-O5	135.56	132.76	133.11	135.24
C3-N4-C9	133.67	143.74	140.36	138.72
C1-S6-C7	111.13	110.69	111.27	111.89
C9-C10-O11	113.54	115.43	118.07	124.09
C9-C10-O12	123.43	121.96	123.98	107.17
O11-C10-O12	123.02	122.613	117.94	128.74
C10-O11-M14	-	140.36	136.88	122.64
C3-O5-M14	-	121.111	123.12	120.42
O5-M14-O11	-	106.260	109.86	117.69
C2-C1-N4-C3	-1.87	0.74	-0.29	-3.04
N4-C1-S6-C7	52.01	59.37	58.52	58.77
C3-N4-C9-C10	44.82	42.47	46.68	55.05
C3-C2-C1-S6	-111.45	-111.6	-110.09	-107.30
C1-C2-C3-O5	175.41	-178.64	-110.09	174.94

Table3: Selected NLMO/NPA bond orders of Ceph and its metal complexes calculated at B3PW91/6-31G** level of theory

	Ceph	$[Ceph-Mn]^+$	$[Ceph-Ni]^+$	$[Ceph-Cu]^+$
C1-C2	0.98	0.97	0.971	0.977
C2-C3	0.95	1.00	1.00	1.00
C1-N4	0.93	0.87	0.87	0.87
C1-S6	0.91	0.96	0.96	0.95
C3-N4	1.14	1.33	1.33	1.04
N4-C9	1.08	0.97	0.97	1.07
C3-O5	1.80	1.35	1.34	1.54
S6-C7	0.92	0.90	0.90	0.91
C7-C8	1.01	1.02	1.02	1.03
C8-C9	1.71	1.68	1.67	1.51
C8-C13	1.04	1.04	1.04	1.06
C9-C10	1.01	1.00	1.02	0.92
C10-C11	1.05	1.04	1.02	1.31
C10-O12	1.71	1.75	1.74	1.57
O5-M14	-	0.19	0.42	0.18
O11-M14	-	0.31	0.66	0.22

Table 4: Total atomic charges for selected atoms described in terms of natural population analysis (NPA) for Ceph and its metal complexes

	Ceph	$[Ceph-Mn]^+$	$[Ceph-Ni]^+$	$[Ceph-Cu]^+$
C1	0.28	0.3	0.3	0.3
C2	-0.56	-0.53	-0.52	-0.54
C3	0.71	0.724	0.725	0.75
N4	-0.47	-0.43	-0.42	-0.46
O5	-0.56	-0.8	-0.69	-0.68
C8	0.08	0.07	0.08	0.09
C10	0.804	0.801	0.79	0.80
O11	-0.705	-0.94	-0.75	-0.79
O12	-0.56	-0.54	-0.5	-0.47
C13	-0.7	-0.7	-0.7	-0.7

Table5. Natural atomic hybrids (NHO), and the polarization coefficient (c_A; in parentheses) for each hybrid in the corresponding NBO of all atoms of β-lactam ring in metal-Ceph complexes calculated at B3PW91/6-31G** level of theory.

Bonds	Hybrides	Ceph	[Ceph-Mn]+	[Ceph-Ni]+	[Ceph-Cu]+
C1-C2	C1	$sp^{2.73}(0.7155)$	$sp^{2.66}(0.7099)$	$sp^{2.7}(0.7093)$	$sp^{2.67}(0.7102)$
	C2	$sp^{3.12}(0.6986)$	$sp^{3.22}(0.7043)$	$sp^{3.22}(0.7049)$	$sp^{3.14}(0.7040)$
C2-C3	C2	$sp^{3.26}(0.7218)$	sp3.42(0.7046)	$sp^{3.48}(0.6994)$	$sp^{3.46}(0.7045)$
	C3	$sp^{1.91}(0.6921)$	$sp^{1.72}(0.7096)$	$sp^{1.68}(0.7147)$	$sp^{1.66}(0.7097)$
C1-N4	C1	$sp^{3.61}(0.6137)$	$sp^{4.06}(0.5898)$	$sp^{4.09}(0.5890)$	$sp^{4.1}(0.5904)$
	N4	$sp^{2.51}(0.7895)$	$sp^{2.88}(0.8076)$	$sp^{2.83}(0.8081)$	$sp^{2.79}(0.8071)$
C3-N4	C3	$sp^{2.26}(0.5933)$	$sp^{2.15}(0.6014)$	$sp^{2.14}(0.6035)$	$sp^{2.37}(0.5978)$
	N4	$sp^{2.11}(0.8050)$	$sp^{1.83}(0.7989)$	$sp^{1.79}(0.7974)$	$sp^{2.21}(0.8016)$
C3-O5	C3	$sp^{1.86}(0.5868)$	$sp^{2.16}(0.5768)$	$sp^{2.25}(0.5738)$	$sp^{2.09}(0.5726)$
	O5	$sp^{1.32}(0.8097)$	$sp^{1.4}(0.8169)$	$Sp\|^{1.36}(0.8190)$	$sp^{1.27}(0.8199)$
N4-C9	N4	$sp^{1.74}(0.7887)$	$sp^{1.68}(0.7997)$	$sp^{1.64}(0.7922)$	$sp^{1.62}(0.7942)$
	C9	$sp^{2.7}(0.6147)$	$sp^{2.89}(0.6004)$	$sp^{2.89}(0.6037)$	$sp^{2.5}(0.6076)$
C9-C10	C9	$sp^{2.2}(0.7258)$	$sp^{1.91}(0.7349)$	$sp^{1.96}(0.7297)$	$sp^{2.02}(0.7420)$
	C10	$sp^{1.64}(.6879)$	$sp^{1.69}(0.6782)$	$sp^{1.68}(0.6838)$	$sp^{1.98}(0.6704)$
C10-O11	C10	$sp^{2.57}(0.5607)$	$sp^{2.52}(0.5595)$	$sp^{2.76}(0.5576)$	$sp^{1.95}(0.5728)$
	O11	$sp^{1.96}(0.8280$	$sp^{1.82}(0.8288)$	$sp^{1.8}(0.8301)$	$sp^{1.45}(0.8197)$
C10-O12(σ)	C10	$sp^{1.99}(0.5858)$	$sp^{1.81}(0.5865)$	$sp^{2.06}(0.5885)$	$sp^{2.04}(0.5849)$
	O12	$sp^{1.44}(0.8104)$	$sp^{1.39}(0.8100)$	$sp^{1.58}(0.8085)$	$sp^{1.66}(0.8111)$
C10-O12(π)	C10	$sp^{99.99}d^{0.4}(0.5625)$	$sp^{100}(0.54)$	$sp^{49.72}d^{0.12}(0.5664)$	sp^{100} (0.5269)
	O12	$sp^{99.99}d^{0.42}(0.8268)$	$sp^{100}(0.8417)$	$sp^{53.5}d^{0.17}(0.8241)$	$sp^{99.99}d^{2.74}(0.8499)$

References

1. Buynak,J.D.;Bachmann,B.;Wu,K.C. *J.Am.Chem. Soc.*116(**1994**)10955-10965.
2. Bulychev,A.;Obrien,M.E.;Massova,I. *J.Am.Chem.Soc.*117(**1995**) 5938-5943.
3. Jackson,P.M.;Roberts,S.M.;Davalli,S.; *J.Chem.Soc.,Perkin Trans.1* (**1996**) 2029-2039.
4. Ghosh,A.;Ghosh,M;Niu,C.;*Chem.Biol.* 3(**1996**)1011-1019.
5. Wladkowski,B.D.;Chenoweth,S.A.;Sanders,J.N. *J.Am.Chem.Soc.*119(**1997**) 6423-6431.
6. Nangia,A.;Chandrakala.P.S.;Balaramakrishna,M.V. *THEOCHEM.* 343(**1995**)157-165.
7. Scottl,D.;"*Antibacterial Chemothrapeutical Agents*".Blakie Academic Publishers.**1997**,Chap3,pp74-158.
8. Anacona,J.R.;Acosta,F. *J.Coor.Chem.*59(**2005**) 621-627
9. CHOHAN Z.H.a, PERVEZ Ha, MOHAMMED KHAN k., RAUF K. and SUPURAN K.T; *Journal of Enzyme Inhibition and Medicinal Chem*istry, 19(1) **(2004)** pp. 51–56
10. HOHAN Z.H., SUPURAN K. T. and SCOZZAFAVA A.; *Journal of Enzyme Inhibition and Medicinal Chemistry*, February Vol. 19 (1) **(2004)**, pp. 79–84
11. HOHAN Z.H.a, SUPURAN K. T. *; Journal of Enzyme Inhibition and Medicinal Chemistry,* October 20(5) **(2005)** 463–468
12. ANACONA J.R. and ACOSTA F.; *Journal of Coordination Chemistry* Vol. 59, No. 6, 15 April **(2005)**, 621–627
13. Chakrawarti,D.B.;Chakrawarti,M.;Maini,P. *J.Indian.Chem.*77(**2000**) 85.
14 Chakrawarti,D.B.;Chakrawarti,M.;Maini,P. *J.Indian.Chem.*77(**2000**) 217.
15 Ming,L.J. "Metallo antibiotics" *Med.Res.Rew.*23(**2003**) 697.
16 Frau,J.;Donoso,J.;Munoz,F.*J.Comp.Chem.*14(**1993**) 1545-1552.
17 Coll. M. , Frau .J. Vilanova B., Donoso J., Mun˜oz F.; Chemical Physics Letters 326(**2000**) 304–310
18 Sadjadi M.S,Farhadyar N,Zare K, *THOCHEM.* 814(2007) 141-145.