See discussions, stats, and author profiles for this publication at: https://www.researchgate.net/publication/235418849

Berberine alkaloid: Quantum chemical study of different forms by the DFT and MP2 methods

ARTICLE in CHEMICAL PHYSICS LETTERS · OCTOBER 2006

Impact Factor: 1.9 · DOI: 10.1016/j.cplett.2006.09.026

CITATIONS

4

READS

72

8 AUTHORS, INCLUDING:



Victor I Danilov

58 PUBLICATIONS 501 CITATIONS

SEE PROFILE



Dmytro Mykolayovych Hovorun

National Academy of Sciences of Ukraine

382 PUBLICATIONS 2,056 CITATIONS

SEE PROFILE



Noriyuki Kurita

Toyohashi University of Technology **144** PUBLICATIONS **1,793** CITATIONS

SEE PROFILE



Anatoly I. Potopalsky

National Academy of Sciences of Ukraine

21 PUBLICATIONS 14 CITATIONS

SEE PROFILE





Chemical Physics Letters 430 (2006) 409-413



Berberine alkaloid: Quantum chemical study of different forms by the DFT and MP2 methods

V.I. Danilov ^a, V.V. Dailidonis ^b, D.M. Hovorun ^a, N. Kurita ^{c,*}, Y. Murayama ^c, T. Natsume ^c, A.I. Potopalsky ^a, L.A. Zaika ^a

^a Institute of Molecular Biology and Genetics, National Academy of Sciences of Ukraine, 150 Zabolotny Street, Kyiv-143, 03143, Ukraine
^b Bogolyubov Institute for Theoretical Physics, National Academy of Sciences of Ukraine, 14b Metrologichaskaya Street, Kyiv-143, 03143, Ukraine
^c Department of Knowledge-based Information Engineering, Toyohashi University of Technology, Tempaku-cho, Toyohashi 441-8580, Japan

Received 31 July 2006; in final form 1 September 2006 Available online 15 September 2006

Abstract

The stable structures and electronic properties for the berberine cation as well as possible ammonium, carbinol and amino-aldehyde forms of protoberberine salts in the presence of hydroxyl ions were investigated by the B3LYP/6-31G(d,p) and MP2/6-31++G(d,p) methods. The geometry optimizations by both methods lead to the nonplanar propeller-twisted and buckled structure for the all forms. The obtained bond lengths and bond angles agree with the experimental values. The comparison of total energies elucidates that the amino-aldehyde form is the most preferable tautomer in gas phase, while the carbinol form is less stable. The least stable tautomer is the ammonium form.

© 2006 Elsevier B.V. All rights reserved.

1. Introduction

Natural alkaloid berberine is mainly contained in plants as hydrochloride or sulfate, and its structural formula is $C_{20}H_{19}NO_5$. Berberine hydrochloride is an isoquinoline alkaloid, and in aqueous solution it prevails as quaternary ammonium cation or quaternary ammonium base 9,10-dimethoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquino[3,2-a]isoquinoline-7-ilium (by the IUAC nomenclature). Actually, it represents a tetra-substituted alkaloid with methylenedioxy group at C2 and C3 positions and two methoxy groups at C9 and C10 positions as shown in Fig. 1.

A structural formula of alkaloid berberine reported in most of publications and monographs is actually a structure of its cation $[C_{20}H_{18}NO_4]^+$. Berberine exhibits antimicrobic action against a great variety of organisms, including bacteria, viruses, fungi, protozoa and clamydia [1,2].

One of the most important molecular targets for antitumor agents is DNA-topoisomerase that forms a covalent bond with both strands of helical DNA by breaking and releasing of sugar-phosphate bonds of DNA backbone. Berberine represents a structural class of organic cations that are important determinants of DNA-topoisomerase inhibition inducing a breakage of complex between this enzyme and DNA (so-called topoisomerase poisoning). The topoisomerase poisoning by certain protoberberine alkaloids is associated with their antitumor effect.

Moreover, berberine was found to possess antitumor properties and the O19 position in berberine analogues (see Fig. 1) is a key determinant of DNA-topoisomerase inhibition. Berberine antitumor activity is attributed to its ability to intercalate into DNA specifically binding to AT-rich regions. Like other intercalating agents, berberine complexation is stipulated by its planar structure that facilitates intercalation and subsequent π -stacking with base pairs. The binding of berberine to DNA untwists double helix of DNA by 11° [3], which is consistent with the intercalation mode of interaction between berberine and DNA.

^{*} Corresponding author. Fax: +81 532 44 6875. E-mail address: kurita@cochem2.tutkie.tut.ac.jp (N. Kurita).

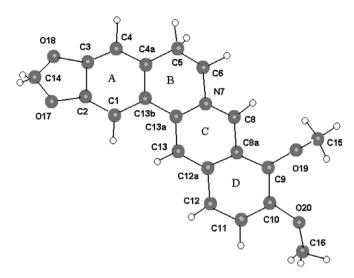


Fig. 1. The structural formula of alkaloid berberine with atom numeration (hydrogen atoms are not numerated).

In addition, computer simulations of protoberberine-DNA complexes supposed that only C and D rings intercalate into DNA, while A and B rings are located outside of internal part of the helix, in the minor groove of DNA [3].

Berberine-containing products proved to be used as effective anti-infectious agents of broad-spectrum for which a minor groove of DNA is a target. Because of increase in bio-terrorism, new anti-infectious compounds of broadspectrum have been found to fight against bioagents such as anthrax (Bacillus anthracis) and smallpox (Variola major), gram-positive bacteria which have AT-rich DNAs (67%). The compounds binding to the AT-rich region of the minor groove are to be effective against these bioagents. This was proved on a new class of compounds brining to small-molecular anti-genomic therapy (SMAT). The investigations of SMAT have been supported by defense agency of advanced research projects (USA) and provided military personnel with easy tool for anthrax and smallpox treatment. SMAT effectively interacts with the minor groove of DNA AT-rich sequences found both in Bacillus anthracis and Variola major. Because of specific mechanisms of action and anti-infectious activity, SMAT was shown to be zeffective against drug-resistant pathogens such as malaria (Plasmodium falciparum) and gram-positive bacterium (for example, Streptococcus pneumoniae). Further elucidation of DNA-binding specificity of SMAT will open up new therapeutic possibilities to apply SMAT in oncology, inflammation, cardiovascular diseases and metabolic disorders.

Thus at present it is proposed that most likely the berberine alkaloid acts as an intercalator during interaction with DNA.

In the presence of hydroxyl ions, quaternary protoberberine salts can exist in three tautomeric forms [4]: ammonium, carbinol and open amino-aldehyde ones. In alkaline solution, berberine salt converts into carbinol form (free base, so-called pseudo-base). In other words, in the presence of anion OH⁻ quaternary protoberberine salts

convert into 8-hydroxyderivatives. The free base is unstable 8-hydroxyadduct and has been studied by means of ¹H and ¹³C NMR spectroscopy. Structures of free bases were also confirmed by mass-spectrometry data. According to the data reported in [5], 8-hydroxyadduct and hydroxide of quaternary berberine alkaloid are supposed to co-exist in equilibrium.

The applied physico-chemical methods did not confirm the amino-aldehyde form (for example, see [6]). Most of investigators therefore do not assume the existence of an aldehyde form in alkaline medium. This does not signify that in alkaline medium this form could not exist at lower concentrations, because many organic reactions indicate the presence of this form. This is explainable by the fact that the aldehyde form may exist in alkaline medium at a very slight concentration and that the equilibrium is constantly maintained in the course of the reaction.

Berberine has been the subject of many experimental studies [1]. However its physico-chemical properties, electronic structure particularly, are not thoroughly investigated so far. Meanwhile, an issue on the electronic structure of berberine cation may be of a great consequence for the structure of its complexes with different biomolecules formed in processes of molecular recognition. Recently the first quantum-chemical work on berberine cation form has appeared [7]. This work reported the bond lengths and angles in cationic form of berberine obtained by the HF/6-31G**, HF/6-311G** and B3LYP/6-311G** methods.

The tautomeric forms of berberine have not been studied theoretically until now, and therefore there is no theoretical data on their structures and electronic properties. Thus, theoretical investigations in gas phase are required not only to obtain the principal data but also to quantitatively determine a relative importance of intermolecular interactions to different structures formed by berberine. In the present study, we investigated the stable structures and electronic properties for the ammonium, carbinol and amino-aldehyde forms of berberine as well as the berberine cation by the density functional theory (DFT) and ab initio MP2 methods.

2. Materials and methods

To elucidate molecular mechanisms of berberine action, we have performed the extensive quantum-chemical investigation on the energetic and structural properties of cationic part of berberine salt as well as the dependence of the obtained results on the method and the basis set used. For this purpose, we used the DFT method with the combined exchange-correlation functionals (PW91, BLYP, and PBEPW91) as well as the hybrid functionals (B3LYP and B3PW91). Furthermore, the calculation for the berberine cation form was performed by more advanced MP2 method. The MP2 calculations were carried out in a frozen frame approximation. For geometry optimization of berberine cation, the standard basis sets 6-31G(d,p) and 6-311G(d,p) were used. Since berberine hydroxide can exist

in solution as an equilibrium mixture of three tautomeric forms, its ammonium, carbinol and amino-aldehyde forms were studied by the MP2/6-31++G(d,p) method. Moreover these tautomeric forms were also studied by the DFT method with B3LYP/6-31G(d,p). The DFT calculations with PW91/6-311G(d,p) were performed using CACHE 5.04 package. At the same for the BLYP/6-311G(d,p), B3LYP/6-311G(d,p), PBEPW91/6-311G(d,p) and B3PW91/6-311G(d,p) calculations, we used PC GAMESS version 7.0 of the GAMESS US quantum chemistry package [8,9]. The main results of our calculations of the berberine cation by MP2/6-31G(d,p) and B3PW91/6-311G(d,p) methods are presented below. The results obtained by both methods are similar each other and moreover they accord with experimental data best of all.

3. Results and discussion

3.1. Stable structure and electronic properties of berberine cation

The bond lengths, propeller twist (PT) angles and dipole moments (μ) of berberine cationic form calculated by MP2/6-31G(d,p) and B3PW91/6-311G(d,p) methods are sum-

marized in Table 1. Table 1 also represents the experimental values for berberine dihydrate bromide [10], berberine azide and berberine thiocyanate [11] obtained by X-ray crystal analysis. It should be noted that the experimental values for some bonds in the berberine salts differ by 0.001–0.029 Å [10–12].

The comparison between the theoretical and experimental data on bond lengths in Table 1 shows a good agreement between them. In the experiments [10,11], the shortest bond in berberine salts was observed for N7–C8 bond in ring C, while the longest bonds are C4a–C5 (sp²–sp³) and C5–C6 (sp³–sp³). Our theoretical results obtained by the MP2 and B3PW91 methods confirm these experimental findings as shown in Table 1.

According to the experimental data, the conformations of cations of all berberine salts are very similar each other. First of all, quaternary cations of berberine consist of hexamerous planar rings in the structure of frame. Deviation from planarity is observed only in the partly saturated ring B. The ring B adopts a twisted half-chair conformation with the atoms C5 and C6 significantly deviated from the best plane of the aromatic rings A and C. In all compounds, the dioxolane ring C2–O17–C14–O18–C3 slightly differs from the planar structure. The structure of berberine

Table 1 The bond lengths (Å), propeller twist (PT) angles and dipole moments (μ) of berberine cation form calculated by (1) MP2/6-31G(d,p) and (2) B3PW91/6-311G(d,p) methods are compared with their experimental values [10,11] for (3) berberine dihydrate bromide, (4) berberine azide, and (5) berberine thiocyanate

Bonds	1	2	3	4	5
C1–C2	1.379	1.369	1.358	_	_
C1-C13b	1.414	1.416	1.409	_	_
C2-C3	1.399	1.396	1.359	_	_
C2-O17	1.366	1.355	1.376	1.380	1.366
C3-C4	1.382	1.376	1.373	_	_
C3-O18	1.364	1.347	1.370	1.379	1.366
C4-C4a	1.403	1.400	1.397	_	_
C4a-C5	1.505	1.505	1.506	1.504	1.506
C4a-C13b	1.410	1.404	1.390	1.400	1.402
C5-C6	1.516	1.515	1.499	1.509	1.500
C6-N7	1.484	1.479	1.485	1.492	1.484
N7-C8	1.339	1.332	1.320	1.334	1.332
N7-C13a	1.388	1.389	1.393	1.391	1.390
C8–C8a	1.400	1.393	1.395	1.398	1.400
C8a-C9	1.416	1.423	1.417	_	_
C8a-C12a	1.436	1.430	1.424	1.423	1.410
C9-C10	1.404	1.397	1.375	_	_
C9-O19	1.360	1.342	1.358	1.376	1.364
C10-C11	1.419	1.414	1.408	_	_
C10-O20	1.349	1.346	1.349	1.358	1.350
C11-C12	1.381	1.375	1.358	_	-
C12-C12a	1.411	1.430	1.400	_	_
C12a-C13	1.404	1.406	1.398	1.404	1.406
C13-C13a	1.387	1.377	1.368	_	_
C13a-C13b	1.463	1.463	1.470	1.464	1.469
C14-O17	1.440	1.430	1.409	1.433	1.433
C14-O18	1.441	1.434	1.439	1.442	1.430
C15-O19	1.449	1.438	1.414	1.443	1.427
C16-O20	1.438	1.425	1.435	1.431	1.436
PT (°)	24	21		10–15	
μ (D)	3.917	3.740	-	_	_

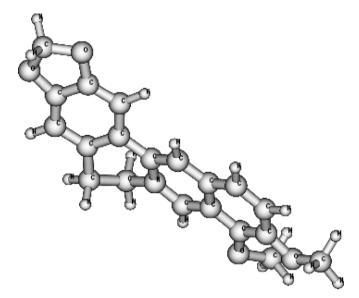


Fig. 2. The stable structure of berberine cationic form obtained by MP2/6-31G(d,p) method.

cation demonstrates that two methoxy groups are oppositely oriented, most likely due to steric constraint. The methoxy group at the position C10 (O20–C16) lies almost in the same plane as the ring D, while the methoxy group at the position C9 (O19–C15) is almost perpendicular to the plane. The calculated results also give evidence that the quaternary cation has relatively planar structure regardless of the theoretical method used. On the whole, the theoretical results obtained by the MP2 and B3PW91 methods are consistent with these experimental data, as shown in Fig. 2.

It is worth noting that in berberine molecule the two aromatic fragments (ring A and fused rings C, D) may be distinguished. According to the experimental data [9–11], the planes of these two fragments are turned one to another by the angle within 10°-15° interval. Meanwhile, in accordance with the results obtained by the MP2/6-31G(d,p) and B3PW91/6-311G(d,p) methods, this angle is 24° and 21°, respectively. Therefore, the theoretically calculated values of the angle noticeably differ from the experimental values. This discrepancy is not surprising, since the calculations were performed for gas phase, while the experimental data were obtained in crystals. In experimentally studied structures, the cations are packed into centrosymmetrical pairs that in turn form the columns parallel to one axis. The space between the columns is filled by anions and water molecules that is evidence of the presence not only of electrostatic forces but also H-bonds in crystals. Therefore, in crystals there are significant packing forces, which affect greatly the spatial structure of berberine cation, resulting in the compact spatial packing.

Thus, the structure of berberine cation obtained by the quantum-chemical investigation gives evidence that rings C and D are most likely to intercalate into DNA helix. This is in agreement with the result of computer simulation [3] for the complexes of protoberberine and DNA.

3.2. Stable structures and electronic properties of berberine tautomers

The structures of three tautomeric forms of berberine (its ammonium, carbinol and amino-aldehyde forms) were optimized in gas phase by the MP2/6-31++G(d,p) method. These optimized structures are shown in Figs. 3–5, respectively. From the comparison of total energies calculated by the MP2/6-31++G(d,p) method, it is elucidated that the most energetically preferable tautomer in gas phase is the amino-aldehyde form shown in Fig. 3. The dipole moment of this amino-aldehyde form is 2.17 D.

The next stable tautomer is a carbinol (non-ionic, prototropic) form, so-called pseudo-base (Fig. 4). This tautomer is less preferable by 19.93 kcal/mol than the most stable one. As seen in Fig. 4, hydrogen bond is formed between the N–H group and the carbonyl oxygen. The pseudo-base has dipole moment of 3.33 D, indicating that this form is stabilized in aqueous solution to the larger degree than the amino-aldehyde one.

The least stable tautomer is an ammonium (ionic, ionotropic) form (Fig. 5), which is less stable by 5.21 kcal/mol than the carbinol one. This fact is not surprising, since the existence of neutral berberine hydroxide is extremely unfavourable in gas phase: one of its fragments is positively charged (ammonium form), while the other is negatively charged (hydroxide). In fact, the initial structure of berberine hydroxide in gas phase could be represented as $[C_{20}H_{18}NO_4]^+[OH^-]$, but its finally optimized structure appeared to be $[C_{20}H_{17}NO_4] \cdot [H_2O]$. In other words, in order that berberine hydroxide does not have localized charges on different fragments, its hydroxyl ion joins a proton of ammonium form and forms a water molecule tightly associated with fragment $[C_{20}H_{17}NO_4]$. This form pos-

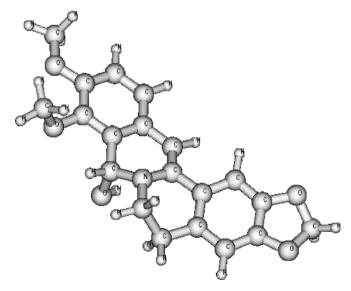


Fig. 3. The stable structure of berberine amino-aldehyde form obtained by MP2/6-31++G(d,p) method.

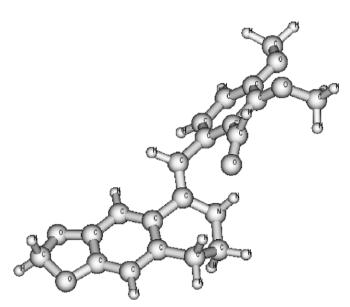


Fig. 4. The stable structure of berberine carbinol form (pseudo-base) obtained by MP2/6-31++G(d,p) method.

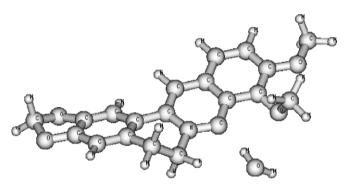


Fig. 5. The stable structure of berberine ammonium form obtained by MP2/6-31++G(d,p) method.

sesses a large dipole moment of 6.22 D, which stabilizes considerably the form in solution.

Although the structure of berberine cation is well known from X-ray crystal analyses [10–12], the structure of the free base of quaternary isoquinoline alkaloid is poorly investigated so far. Some experimental data on the structure of the berberine free base were reported in [13]. The comparison of the calculated values of bond lengths and angles in berberine pseudo-base (8-hydroxydihydroberberine) and the experimental values reveals a good agreement. All bond lengths and angles of 8-hydroxydihydroberberine are within usual values. The calculated length of C8-OH bond is 1.436 Å, while the experimental value is 1.440 Å. The theoretical magnitude of the sum of three valence angles around nitrogen atom is 359.99°, while the experimental value equals to 353.98°. This implies that the hybridisation around nitrogen atom is close to sp² hybridisation. Partly hydrogenated hetero-ring B adopts a conformation of twisted half-chair, while the ring C has a conformation of shallow half-chair. The C8 atom is deviated by 0.373 Å from the plane made by the rings C and D. The hemiaminoacetal hydroxyl group C8–OH is located at the axial position as to the ring C conformation as shown in Fig. 4. The methoxy group attached to C9 atom is almost perpendicular to the D plane, while the other methoxy group attached to C10 atom is almost in the plane. The calculated torsion angles are 117.0° [C8a-C9-O19–C15], -66.7° [C10–C9–O19–C15], -176.7° [C9– C10-O20-C16] and 1.8° [C11-C10-O20-C16], respectively. The corresponding experimental values are 117.7°, 71.3°, 175.7° and -3.6° . Therefore, the theoretical data fully agree with the experimental ones [13]. Similar to quaternary berberine salts, this is a general feature of berberine quaternary 8-adducts. Within a reasonable accuracy, the dioxolane ring C2-C3-O18-C14-O17 is planar with the average deviation from the plane of 0.0511 Å. The angle between two aromatic rings A and D is equal to 148.5°. There is a good agreement between this angle and its experimental magnitude.

4. Conclusions

The present quantum-chemical investigations based on MP2 and DFT methods for the tautomeric forms of berberine alkaloid as well as its cationic form proved to obtain a comparable structural parameters to the experimentally observed ones. In spite of the nonplanar propeller-twisted and buckled structures for all berberine alkaloid forms, two rings C and D in the cation remain coplanar structure that is favored for the intercalation into DNA helix.

Acknowledgements

V.I. Danilov thanks Dr. Alex A. Granovsky (Moscow State University) for providing PC GAMESS version 7.0. We are grateful to the reviewer for the useful remarks.

References

- A.I. Potopalsky, L.I. Petlichnaya, S.I. Ivasivka, Modification of Berberine Alkaloid, Naukova dumka, Kiev, 1982.
- [2] T.C. Birdsal, G.S. Kell Berberine, Alt. Med. Rev. 2 (1997) 94.
- [3] T.-K. Li et al., Biochemistry 39 (2000) 7107.
- [4] D. Beke, in: A.R. Katritzy (Ed.), Advances in Heterocyclic Chemistry, vol. 1, Academic Press, New York, 1963.
- [5] R. Marek, P. Sečkárová, D. Hulová, J. Marek, J. Dostál, V. Sklenáŕ, J. Nat. Prod. 66 (2003) 481.
- [6] V. Šimánek, V. Preininger, S. Hegerová, F. Šantavy, Coll. Czech. Chem. Commun. 37 (1972) 2746.
- [7] M.-J. Huang, K.S. Lee, S.J. Harley, Int. J. Quant. Chem. 105 (2005) 396
- [8] M.W. Schmidt et al., J. Comput. Chem. 14 (1993) 1347.
- [9] A.A. Granovsky, PC Gamess, version 7.0, Moscow State University, Moscow, Russia, 2006.
- [10] B.M. Kariuki, W. Jonson, Acta Cryst. C51 (1995) 1234.
- [11] S. Man, M. Potáček, M. Nečas, Z. Žák, J. Dostál, Molecules 6 (2001)
- [12] G. Blasko, G.A. Cordell, S. Bhamarapravati, C.W.W. Beecher, Heterocycles 27 (1988) 911.
- [13] J. Dostál et al., J. Mol. Struct. 687 (2004) 135.