ORIGINAL ARTICLE

AB INITIO STUDY OF THE Na–COLCHICINE POSITIVELY CHARGED COMPLEX

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Abstract

Colchicine's multimodal mechanism of action, strongly correlated with its inherent chemical properties, is still in the focal point of the very recent biomedical studies. In the present study we investigated the geometry and electronic structure of Na–colchicine positively charged complex (metal adduct ion) using DFT and Hartree-Fock calculations. The geometry of the metal adduct is investigated for eight different positions of sodium ion relative to the colchicine molecule. The influence of Na^+ upon the electronic structure of colchicine is emphasized by pointing out the changes in the total energy and electronic structure of the resulting complex.

Rezumat

Mecanismul de acţiune multimodal al colchicinei, fenomen strâns corelat cu proprietăţile chimice ale acesteia, beneficiază în continuare de o atenţie sporită în cercetările biomedicale foarte recente. În studiul de faţă s-a investigat geometria şi structura electronică a complexului cationic (aduct metalic) Na-colchicină prin calcule DFT şi Hartree-Fock. Geometria aductului metalic a fost investigată pentru opt poziţii distincte ale ionului de sodiu faţă de molecula colchicinei. Influenţa Na^+ asupra structurii electronice a colchicinei a fost pusă în evidenţă prin urmărirea modificărilor energiei totale şi a structurii electronice a complexului rezultat.

Keywords: colchicine, metal adduct ion, DFT, electronic structure

Introduction

Colchicine (CC), (S)-N-(5,6,7,9-tetrahydro-1,2,3,10-tetramethoxy-9--oxobenzo-(a(-heptalen-7-yl)-acetamide, is the main protoalkaloid of the poisonous plant *Colchicum autumnale L.* (meadow saffron). CC has been used for centuries in the treatment of acute gouty arthritis, but in the last decade, it has also proven its efficiency in the treatment of other inflammatory diseases such as, Mediterranean fever, Behçet's syndrome, osteoarthritis, liver cirrhosis and various cardiovascular diseases [10]. Selectively binding to tubulin, a structural glycoprotein of the mitotic spindle, CC shows a strong antimitotic activity [1], being a model molecule for new chemotherapeutic agents development. Nevertheless, there are still many unknowns related to its therapeutic use, reporting also cognitive impairment and development of sporadic Alzheimer's disease upon its central administration [6, 7]. It is therefore justified the great interest towards the physico-chemical characterization of CC and its complexes.

The molecule of CC contains a trimethoxybenzene moiety (ring A), a seven-membered ring with an acetamido group in C-7 position (ring B) and a methoxytropolone moiety (ring C). Having an asymmetric carbon atom (C-7) associated with an axial atropisomerism, CC has four enantiomers, among which (-)-(aS,7S)-colchicine (Figure 1) is the natural congener [3].

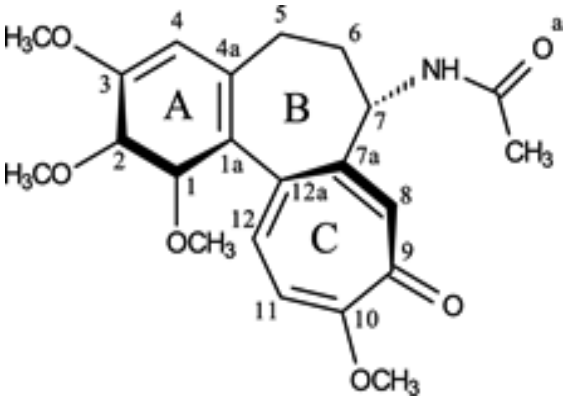

Figure 1.
Structure of (-)-(aS,7S)-colchicine

Formation of stable complexes of 1:1 stoichiometry between CC and alkali metal cations (Li^+, Na^+, K^+)

with potential biological role were reported by Kurek et al. [8]. The most stable structures were found to be those in which the acetamide group is involved in the coordination process. Moreover, alkali metal adducts of CC are also encountered in the mechanistic study of its redox behaviour using electrochemistry coupled to mass spectrometry [2]. In assays performed in non-protogenic media (i.e. dimethylformamide) these metal adduct ions of CC become the base peak of the mass spectra, manifesting a considerable stability towards tandem MS fragmentation.

Such stable cationic complexes of CC in solution might also have an influence on the pharmacokinetics and pharmacodynamics of CC molecule. Moreover, the stability and stoichiometry of these complexes may have a crucial influence on CC's analytical properties, enabling novel approaches for its spectroscopic analysis [5, 14], whereas the understanding of the nature of binding energies may open the way for future pharmaceutical applications of such complexes.

In the present paper a detailed computational analysis of the geometry and electronic structure of Na–CC positively charged complex is presented. The study is based on DFT (Density Functional Theory) calculation. This strategy has been proved as effective in the study of interaction between organic molecules and metal atoms [9]. Our aim was to find the most probable geometrical structure of the Na^+–CC complex, and to give a detailed quantum-mechanical description of the factors leading to this structure. As far as we know, this study represents the first report on ab initio computational analysis for Na-colchicine positively charged complex, the previous computational results on colchicine complexes with lithium, sodium and potassium salts, reported by Kurek et al. [8], were obtained by using PM5 semi empirical calculations.

Materials and Methods

Computational details

The geometrical structure of the CC molecule was built up using Molden [4]. In the first part of the paper a periodic DFT study is presented, for which the SIESTA package was used [11, 13]. The whole system was confined into a cubic periodic cell (a = 20 Å) which is sufficiently large to avoid the interaction between the periodical images of the molecule. A standard double-zeta polarized (DZP) basis set with an energy shift of 50 meV was used. The presence of polarization functions and the small cut-off (300 Ry) ensures for a good description of the molecule-ion interactions. For all atoms Troullier-Martins pseudo-potentials [15] and Perdew-Burke-Ernzerhof (PBE) version of Generalized Gradient Approximation (GGA) as exchange-correlation functional [12] were use.

Results and Discussion

a) Investigation of free colchicine. In the first step the structure of CC, was relaxed up to a gradient of 10^{-4} Ha/Bohr. The same procedure was applied to the structures of CC^+ and CC^- ions. In order to get a first idea of the changes induced in the electronic structure by the ionization process, the charge density of the ionized molecule ($\rho^{\pm 1}$) was subtracted from that of the neutral one(ρ^0). The variation of the total electronic density is

$$\Delta\rho^{\pm} = \rho^0 - \rho^{\pm 1}.$$

The contour plots of $\Delta\rho^{\pm}$ are given in Figure 2. It can be seen that the gain of one electron influences the states located on the heptacycle C (Figure 2, left). On the other hand, the loss of one electron influences states located practically over the whole molecule (Figure 2, right). These results give a qualitative indication of the possible regions where a charge transfer between CC and Na may occur.

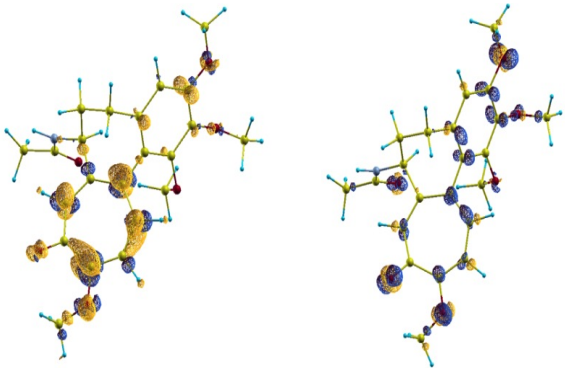

Figure 2.
Contour plot of $\Delta\rho^{\pm}$ (see text for definition) for $\rho^0_{CC} - \rho^-_{CC}$ (left) and $\rho^0_{CC} - \rho^+_{CC}$ (right).
Blue: positive values, yellow: negative values

b) Investigation of Na–CC positively charged system. In order to find the most probable geometrical structure of the Na–CC positively charged complex, eight different positions of Na^+ in the proximity of CC molecule were taken into account (Table 1). For each of these structures, the initial system geometry was relaxed up to a maximum gradient of 0.04 eV/Bohr.

Table I

The positions of Na^+ cation relative to CC molecule before and after the structural relaxation
The different geometries are distinguished by indicating the distances D (in Å) between Na^+ and selected atoms (X) in CC molecule, before (values in brackets) and after relaxation.
For Ors. 1, 2 and 7, these atoms are: A = C_1, B = C_2, C = C_3, D = O_1, E = O_2, F = O_a, G = O_9, H = O_{10}.
For Ors. from 3 to 6 and Or. 8, A = C_8, B = C_9, C = C_{10}, columns D–H denoting the same atoms.
For atoms notations, see Figure 1.

D [Na^+–X] [Å]								
Or./ Atom	A	B	C	D	E	F	G	H
1	2.9 (2.0)	2.7 (2.1)	3.0 (2.2)	3.7 (3.0)	3.3 (3.1)	2.2 (3.9)	7.9 (7.4)	8.3 (7.7)
2	2.7 (2.0)	2.9 (2.1)	2.9 (2.2)	3.6 (3.0)	3.9 (3.3)	6.9 (6.1)	7.3 (7.3)	7.0 (7.0)
3	3.6 (2.0)	4.3 (2.2)	4.0 (2.1)	2.5 (3.9)	5.1 (6.5)	2.2 (3.3)	5.5 (3.4)	5.0 (3.2)
4	4.2 (2.0)	3.3 (1.9)	4.1 (2.1)	8.9 (4.7)	11.3 (6.9)	7.4 (5.0)	2.1 (2.9)	4.1 (3.3)
5	8.9 (9.6)	10.0 (10.5)	9.8 (10.2)	4.8 (5.5)	2.2 (2.8)	7.2 (8.5)	11.2 (11.7)	11.0 (11.4)
6	7.4 (7.2)	8.1 (7.8)	7.4 (7.1)	2.3 (2.1)	2.3 (2.1)	6.0 (6.0)	9.3 (9.0)	8.3 (8.0)
7	3.2 (7.2)	4.5 (8.6)	5.4 (9.6)	2.5 (6.4)	5.1 (9.1)	2.2 (6.9)	5.0 (4.6)	5.4 (2.0)
8	4.0 (4.2)	3.3 (3.3)	4.4 (4.2)	9.0 (9.0)	11.2 (11.4)	7.0 (7.3)	2.1 (2.0)	4.6 (4.2)

Orientation (Or.) 1: Na^+ ion is positioned initially above ring A (Figure 1), on the same side of the plane with O_a. After relaxation, distances between Na^+ and C_1, C_2 atoms increase until 2.9 Å, respectively 2.7 Å, as Na^+ is attracted towards O_a ($Na^+\cdots O_a$ distance after relaxation is 2.2 Å compared with the initial value, 3.9 Å). Or. 2: Na^+ ion is positioned below ring A, on its symmetry axis. The initial distances between Na^+ and the atoms belonging to A ring are about the same as for Or. 1. In the final position, Na^+ is slightly pushed away from ring A along the same symmetry axis. Or. 3: Na^+ ion is positioned above the methyltropolonic ring C, on the same side of the plane with O_a. After relaxation, Na^+ is pushed away from C_8, C_9 and C_{10} and from O_9 and O_{10} as well. Also, torsions of the simple bonds of N and C_1–O_1 were noted. Or. 4: Na^+ ion is positioned below ring C on a median direction orthogonal on methyltropolonic ring C. In the relaxed structure, Na^+ is pushed away from the centre of C ring and it is directed towards the carbonilic oxygen O_9, passing close by C_9. As a consequence of pushing also from ring A, the distances $Na^+\cdots O_1$, O_2 and O_3 increase. Or. 5: Na^+ is positioned slightly below plane of the benzene ring, between O_2 and O_3. In the final position, Na^+ comes above this plane, being pushed very slightly from O_3 and towards O_1 and O_2. Or. 6: Na^+ is positioned in the plane formed by O_1, O_2 and O_3, between O_1 and O_2, very slightly below ring A; the final position of Na^+ is slightly above ring's A plane. Na^+ is not significantly displaced from the initial position after relaxation. Or. 7: Shows the most spectacular behaviour of Na^+. Initially it is positioned above the plane of C, between O_{10} and C_{11}. After relaxation Na^+ is strongly attracted by O_1, O_2, O_3 and O_a (Figure 3) (and C_1, C_2 and C_3), being pushed from O_9 and O_{10}. Or. 8: Na^+ is positioned on C_9–O_9 bond's axis at 2.0 Å from O_9. In the final structure, this distance increases slightly (2.1 Å), Na^+ getting slightly below plane C.

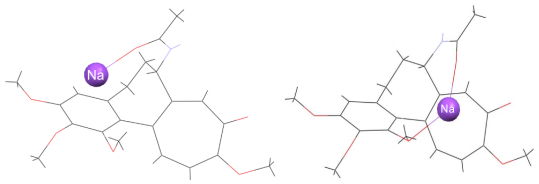

Figure 3.
The most stable relaxed geometries of CC molecule having in proximity a Na^+ ion, Or. 1 (left) and Or. 3 (right)

It can be observed that Or. 3 and Or. 7 lead to the same final structure after relaxation (Table 1, selected geometric parameters presented in bold). All other initial models lead to different stable geometries of the CC–Na ionic complex. In order to quantitatively define the concept of "stable complexes", two types of binding energy were considered. First, the complex formed from Na^+ and CC as component units, whereas in the second case, from Na and CC^+ were considered, being expressed as

$$\Delta E_1 = E_{(Na-CC)^+} - E_{Na^+} - E_{CC}$$

And

$$\Delta E_2 = E_{(Na-CC)^+} - E_{Na} - E_{CC^+}.$$

For each complex resulting after relaxation the values of ΔE_1 and ΔE_2 were computed. By inspecting the results (Table 2), it can be noted that Or. 3 and Or. 7 have the lowest total energies. Or. 1 with a slightly smaller binding energy (i.e. a difference of about 0.2 eV compared with Or. 3 and Or. 7) may also be formed. The latter structure is very similar with the one reported for Li^+–CC complex [8].

Table II

Binding energy (ΔE) for Na^+–CC and Na–CC^+ ionic complexes, for each of the eight positions investigated

ΔE [eV]								
System/Orientation	1	2	3	4	5	6	7	8
Na^+–CC	-2.02	-1.17	-2.29	-1.74	-1.60	-1.62	-2.27	-1.73
Na–CC^+	-3.71	-2.86	-3.98	-3.43	-3.29	-3.31	-3.96	-3.42

Further on, the investigation of the electronic structure of the most stable complexes (Or. 1 and Or. 3) was considered by the analysis of the projected density of states (PDOS) onto the atomic orbitals of the system's components. The PDOS summed over the atoms of CC^+ molecule was compared with those of the CC in the Na–CC^+ system (Figure 4). It can be seen that the HOMO-LUMO gap of the free CC^+ molecule is shifted to smaller energies when the interaction with Na occurs, however the value of this gap is roughly unchanged. On the other hand, the PDOS below this gap is strongly influenced by Na, both for Or. 1 and Or. 3. This clearly shows that the nature of the interaction is different in the two cases. Moreover, for Or. 1 the shift is comparable with the binding energy (about 2.3 eV), while this is not the case for Or. 3, where the shift is smaller (about 1.0 eV).

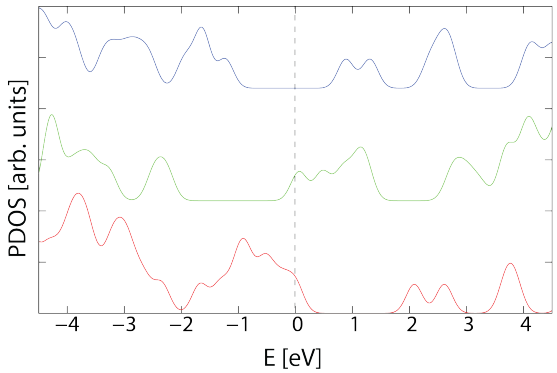

Figure 4.

PDOS of the isolated CC^+ molecule (bottom) and in proximity of Na^+ ion for Or. 1 (middle) and Or. 3 (top). Fermi level was set to zero.

Conclusions

The geometry and electronic structure of Na–CC positively charged complexes (metal adduct ions) by using *ab initio* methods were investigated. The DFT structural relaxation shows that the most stable geometry of the complex is attained when the Na^+ ion is located above the methyltropolonic ring. A second geometry with slightly lower binding energy was also found.

Acknowledgements

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