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Cholera toxin time-resolved absorption and resonance FT-IR and raman biospectroscopy and density functional theory (DFT) investigation of vibronic-mode coupling structure in vibrational spectra analysis

Alireza Heidari^{1,2*}, Jennifer Esposito¹ and Angela Caissutti¹

¹Faculty of Chemistry, California South University, 14731 Comet St. Irvine, CA 92604, USA ²American International Standards Institute, Irvine, CA 3800, USA

Abstract

Cholera toxin (also known as choleragen and sometimes abbreviated to CTX, Ctx or CT) is AB5 multimeric protein complex secreted by the bacterium Vibrio cholerae. CTX is responsible for the massive, watery diarrhea characteristic of cholera infection. It is a member of the Heat-labile enterotoxin family. Parameters such as FT-IR and Raman vibrational wavelengths and intensities for single crystal Cholera Toxin are calculated using density functional theory and were compared with empirical results. The investigation about vibrational spectrum of cycle dimers in crystal with carboxyl groups from each molecule of acid was shown that it leads to create Hydrogen bonds for adjacent molecules. The current study aimed to investigate the possibility of simulating the empirical values. Analysis of vibrational spectrum of Cholera Toxin is performed based on theoretical simulation and FT-IR empirical spectrum and Raman empirical spectrum using density functional theory in levels of HF/6-31G*, HF/6-31++G**, MP2/6-31G, MP2/6-31++G**, BLYP/6-31G, BLYP/6-31++G**, B3LYP/6-31G and B3LYP6-31-HEG**. Vibration modes of methylene, carboxyl acid and phenyl cycle are separately investigated. The obtained values confirm high accuracy and validity of results obtained from calculations.



Molecular structure of Cholera Toxin [1-42].

*Correspondence to: Alireza Heidari, Faculty of Chemistry, California South University, 14731 Comet St. Irvine; American International Standards Institute, Irvine, CA 3800, USA, E-mail: Scholar.Researcher.Scientist@gmail.com; Alireza.Heidari@calsu.us; Central@aisi-usa.org

Key words: Vibronic Structure, Vibrational Spectra Analysis, Density Functional Theory (DFT), Cholera Toxin, Non-Focal Functions of Becke, Correlation Functions of Lee-Yang-Parr, Time-Resolved Absorption and Resonance, FT-IR and Raman Biospectroscopy

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Introduction

Cholera toxin (also known as choleragen and sometimes abbreviated to CTX, Ctx or CT) is AB5 multimeric protein complex secreted by the bacterium Vibrio cholerae. CTX is responsible for the massive, watery diarrhea characteristic of cholera infection. It is a member of the Heat-labile enterotoxin family. Density Functional Theory (DFT) is one of the most powerful calculation methods for electronic structures [5-7]. Numerous results have been previously studied and indicate successful use of these methods [8-10]. The theory is one of the most appropriate methods for simulating the vibrational wavenumbers, molecular structure as well as total energy. It may be useful to initially consider the calculated results by density functional theory using HF/6-31G*, HF/6-31++G**, MP2/6-31G, MP2/6-31++G**, BLYP/6-31G, BLYP/6-31++G**, B3LYP/6-31G and B3LYP6-31-HEG** approach [11-16]. It should be noted that calculations are performed by considering one degree of quantum interference as well as polarization effects of 2d orbitals in interaction [17-364].

Details of calculations

All calculations of molecular orbital in the base of ab are performed by Gaussian 09. In calculation process, the structure of Cholera Toxin molecule (Figure 1) is optimized and FT-IR and Raman wavenumbers are calculated using HF/6-31G*, HF/6-31++G**, MP2/6-31G, MP2/6-31++G**, BLYP/6-31G, BLYP/6-31++G**, B3LYP/6-31G and B3LYP6-31-HEG** base. All optimized structures are adjusted with minimum energy. Harmonic vibrational wavenumbers are calculated using second degree of derivation to adjust convergence on potential surface as good as possible and to evaluate vibrational energies at zero point. In optimized structures considered in the current study, virtual frequency modes are not observed which indicates that the minimum potential energy surface is correctly chosen. The optimized geometry is calculated by minimizing the energy relative to all geometrical quantities without forcing any constraint on molecular symmetry. Calculations were performed by Gaussian 09. The current calculation is aimed to maximize structural optimization using density functional theory. The calculations of density functional theory are performed by HF/6-31G*, HF/6-31++G**, MP2/6-31G, MP2/6-31++G**, BLYP/6-31G, BLYP/6-31++G**, B3LYP/6-31G and B3LYP6-31-HEG** function in which non-focal functions of Becke and correlation functions of Lee-Yang-Parr beyond the Franck-Condon approximation are used. After completion of optimization process, the second order derivation of energy is calculated as a function of core coordination and is investigated to evaluate whether the structure is accurately minimized. Vibrational frequencies used to simulate spectrums presented in the current study are derived from these second order derivatives. All calculations are performed for room temperature of 373 (K).

Vibration analysis

Analysis of vibrational spectrum of Cholera Toxin is performed based on theoretical simulation and FT–IR empirical spectrum and Raman empirical spectrum using density functional theory in levels of HF/6–31G*, HF/6–31++G**, MP2/6–31G, MP2/6–31++G**, BLYP/6– 31G, BLYP/6–31++G**, B3LYP/6–31G and B3LYP6–31–HEG**. Vibration modes of methylene, carboxyl acid and phenyl cycle are separately investigated.

C–H stretching vibrations in single replacement of benzene cycles are usually seen in band range of $3150-3400 \text{ cm}^{-1}$. Weak Raman bands are at 3139 cm^{-1} and 3152 cm^{-1} . C–C stretching mode is a strong



Figure 1. Different sections of the Cholera Toxin [43-93]

Raman mode at 1149 cm⁻¹. Raman weak band is seen at 1623 cm⁻¹, too. Bending mode of C–H is emerged as a weak mode at 1348 cm⁻¹ and 1147 cm⁻¹ and a strong band at 1231 cm⁻¹ in Raman spectrum. Raman is considerably active in the range of 1150–1400 cm⁻¹ which 1153 cm⁻¹ indicates this issue.

C-H skew-symmetric stretching mode of methylene group is expected at 3135 cm⁻¹ and its symmetric mode is expected at 2949 cm⁻¹. Skew-symmetric stretching mode of CH₂ in Cholera Toxin has a mode in mid-range of Raman spectrum at 3050–3170 cm⁻¹. When this mode is symmetric, it is at 3045 cm⁻¹ and is sharp. The calculated wavenumbers of higher modes are at 3013 cm⁻¹ and 3043 cm⁻¹ for symmetric and skew-symmetric stretching mode of methylene, respectively.

Scissoring vibrations of CH₂ are usually seen at the range of 1477–1531 cm⁻¹ which often includes mid–range bands. Weak bands at 1490 cm⁻¹ are scissoring modes of CH₂ in Raman spectrum. Moving vibrations of methylene are usually seen at 1469 cm⁻¹. For the investigated chemical in the current study, these vibrations are at 1289 cm⁻¹ were calculated using density functional theory. Twisting and rocking vibrations of CH₂ are seen in Raman spectrum at 865 cm⁻¹ and 1139 cm⁻¹, respectively, which are in good accordance with the results at 849 cm⁻¹ and 1114 cm⁻¹, respectively.

In a non-ionized carboxyl group (COOH), stretching vibrations of carbonyl [C=O] are mainly observed at the range of 1790–1838 cm⁻¹. If dimer is considered as an intact constituent, two stretching vibrations of carbonyl for symmetric stretching are at 1690–1735 cm⁻¹ in Raman spectrum. In the current paper, stretching vibration of carbonyl mode is at 1747 cm⁻¹ which is a mid–range value.

Stretching and bending bands of hydroxyl can be identified by width and band intensity which in turn is dependent on bond length of Hydrogen. In dimer form of Hydrogen bond, stretching band of O–H is of a strong Raman peak at 1317 cm⁻¹ which is due to in–plain metamorphosis mode. Out–of–plain mode of O–H group is a very strong mode of peak at 999 cm⁻¹ of Raman spectrum. The stretching mode of C–O (H) emerges as a mid–band of Raman spectrum at 1197 cm⁻¹.

Lattice vibrations are usually seen at the range of 0–1050 cm⁻¹. These modes are induced by rotary and transferring vibrations of molecules and vibrations and are including Hydrogen bond. Bands with low wavenumbers of Hydrogen bond vibrations in FT–IR and Raman spectrum (Figure 2) are frequently weak, width and unsymmetrical. Rotary lattice vibrations are frequently stronger than transferring ones. Intra-molecular vibrations with low wavenumbers involving two-bands O–H …O dimer at 588 cm⁻¹, 693 cm⁻¹ and 749 cm⁻¹ are attributed to a rotary moving of two molecules involving in–plain rotation of molecules against each other.

Conclusion and summary

Calculations of density functional theory using HF/6–31G*, HF/6– 31++G**, MP2/6–31G, MP2/6–31++G**, BLYP/6–31G, BLYP/6– 31++G**, B3LYP/6–31G and B3LYP6–31–HEG** levels were used to obtain vibrational wavenumbers and intensities in single crystal of Cholera Toxin. Investigation and consideration of vibrational spectrum confirm the formation of dimer cycles in the investigated crystal with carboxyl groups from each Hydrogen molecule of acid protected from adjacent molecules. The calculated vibrational spectrum which obtains from calculations of density functional theory is in good accordance with recorded empirical values which indicates successful simulation



Figure 2. 3D Simulation of (a) FT-IR spectrum and (b) Raman spectrum of Cholera Toxin

of the problem. The obtained results indicate that the results obtained from theoretical calculations are valid through comparing with empirical recorded results.

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