

# Cobrotoxin 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

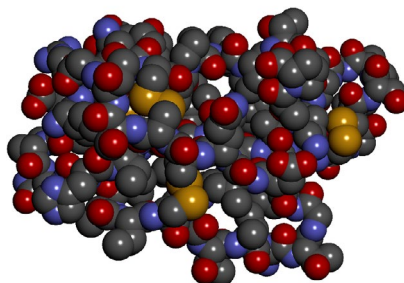
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## Abstract

$\alpha$ -Cobrotoxin is a substance of the venom of certain *Naja* cobras. It is a nicotinic acetylcholine receptor (nAChR) antagonist which causes paralysis by preventing the binding of acetylcholine to the nAChR. Parameters such as FT-IR and Raman vibrational wavelengths and intensities for single crystal Cobrotoxin 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 Cobrotoxin 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<sup>\*</sup>, HF/6-31++G<sup>\*\*</sup>, MP2/6-31G, MP2/6-31++G<sup>\*\*</sup>, BLYP/6-31G, BLYP/6-31++G<sup>\*\*</sup>, B3LYP/6-31G and B3LYP6-31-HEG<sup>\*\*</sup>. 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 Cobrotoxin.

## Introduction

$\alpha$ -Cobrotoxin is a substance of the venom of certain *Naja* cobras. It is a nicotinic acetylcholine receptor (nAChR) antagonist which causes paralysis by preventing the binding of acetylcholine to the nAChR. 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<sup>\*</sup>, HF/6-31++G<sup>\*\*</sup>, MP2/6-31G, MP2/6-31++G<sup>\*\*</sup>, BLYP/6-31G, BLYP/6-31++G<sup>\*\*</sup>, B3LYP/6-31G and B3LYP6-31-HEG<sup>\*\*</sup> 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 Cobrotoxin molecule (Figure 1) is optimized and FT-IR and Raman

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**Key words:** vibronic structure, vibrational spectra analysis, density functional theory (dft), cobrotoxin, 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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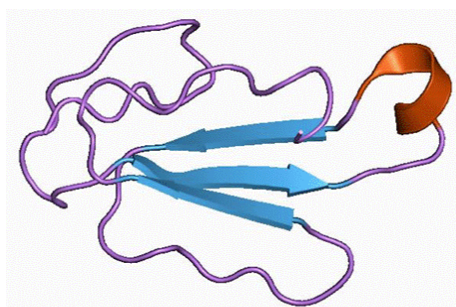


Figure 1. Section of the cobrotoxin

wavenumbers are calculated using HF/6-31G<sup>\*</sup>, HF/6-31++G<sup>\*\*</sup>, MP2/6-31G, MP2/6-31++G<sup>\*\*</sup>, BLYP/6-31G, BLYP/6-31++G<sup>\*\*</sup>, B3LYP/6-31G and B3LYP6-31-HEG<sup>\*\*</sup> 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<sup>\*</sup>, HF/6-31++G<sup>\*\*</sup>, MP2/6-31G, MP2/6-31++G<sup>\*\*</sup>, BLYP/6-31G, BLYP/6-31++G<sup>\*\*</sup>, B3LYP/6-31G and B3LYP6-31-HEG<sup>\*\*</sup> 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 444 (K).

## Vibration Analysis

Analysis of vibrational spectrum of Cobrotoxin 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<sup>\*</sup>, HF/6-31++G<sup>\*\*</sup>, MP2/6-31G, MP2/6-31++G<sup>\*\*</sup>, BLYP/6-31G, BLYP/6-31++G<sup>\*\*</sup>, B3LYP/6-31G and B3LYP6-31-HEG<sup>\*\*</sup>. 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 2900-3150 cm<sup>-1</sup>. Weak Raman bands are at 2889 cm<sup>-1</sup> and 2902 cm<sup>-1</sup>. C-C stretching mode is a strong Raman mode at 799 cm<sup>-1</sup>. Raman weak band is seen at 1273 cm<sup>-1</sup>, too. Bending mode of C-H is emerged as a weak mode at 998 cm<sup>-1</sup> and 797 cm<sup>-1</sup> and a strong band at 881 cm<sup>-1</sup> in Raman spectrum. Raman is considerably active in the range of 800-1050 cm<sup>-1</sup> which 793 cm<sup>-1</sup> indicates this issue.

C-H skew-symmetric stretching mode of methylene group is expected at 785 cm<sup>-1</sup> and its symmetric mode is expected at 2599 cm<sup>-1</sup>. Skew-symmetric stretching mode of CH<sub>2</sub> in Cobrotoxin has a mode in mid-range of Raman spectrum at 2700-2820 cm<sup>-1</sup>. When this mode is symmetric, it is at 2795 cm<sup>-1</sup> and is sharp. The calculated wavenumbers of higher modes are at 2663 cm<sup>-1</sup> and 2693 cm<sup>-1</sup> for symmetric and skew-symmetric stretching mode of methylene, respectively.

Scissoring vibrations of CH<sub>2</sub> are usually seen at the range of 1127-1181 cm<sup>-1</sup> which often includes mid-range bands. Weak bands at 1140 cm<sup>-1</sup> are scissoring modes of CH<sub>2</sub> in Raman spectrum. Moving vibrations of methylene are usually seen at 1069 cm<sup>-1</sup>. For the investigated chemical in the current study, these vibrations are at 939 cm<sup>-1</sup> were calculated using density functional theory. Twisting and rocking vibrations of CH<sub>2</sub> are seen in Raman spectrum at 615 cm<sup>-1</sup> and 789 cm<sup>-1</sup>, respectively, which are in good accordance with the results at 499 cm<sup>-1</sup> and 764 cm<sup>-1</sup>, respectively.

In a non-ionized carboxyl group (COOH), stretching vibrations of carbonyl [C=O] are mainly observed at the range of 1440-1488 cm<sup>-1</sup>. If dimer is considered as an intact constituent, two stretching vibrations of carbonyl for symmetric stretching are at 1340-1385 cm<sup>-1</sup> in Raman spectrum. In the current paper, stretching vibration of carbonyl mode is at 1397 cm<sup>-1</sup> 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 967 cm<sup>-1</sup> which is due to in-plane metamorphosis mode. Out-of-plane mode of O-H group is a very strong mode of peak at 649 cm<sup>-1</sup> of Raman spectrum. The stretching mode of C-O (H) emerges as a mid-band of Raman spectrum at 847 cm<sup>-1</sup>.

Lattice vibrations are usually seen at the range of 0-1000 cm<sup>-1</sup>. 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

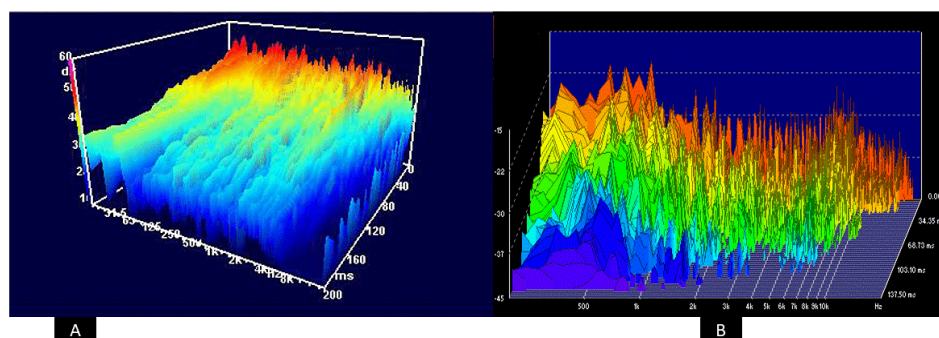


Figure 2. 3D Simulation of (a) FT-IR spectrum and (b) Raman spectrum of cobrotoxin

ones. Intra-molecular vibrations with low wavenumbers involving two-bands O-H ...O dimer at 688 cm<sup>-1</sup>, 693 cm<sup>-1</sup> and 649 cm<sup>-1</sup> 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 Cobrotoxin. 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 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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