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Tetrodotoxin (TTX) 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 Esposito[1], Angela Caissutti[1]

[1]Faculty of Chemistry, California South University, 14731 Comet St. Irvine, CA 92604, USA

[2]American International Standards Institute, Irvine, CA 3800, USA

Abstract

Tetrodotoxin (TTX) is a potent neurotoxin. Its name derives from Tetraodontiformes, an order that includes pufferfish, porcupinefish, ocean sunfish, and triggerfish; several of these species carry the toxin. Although tetrodotoxin was discovered in these fish and found in several other aquatic animals (e.g., in blue–ringed octopuses, rough–skinned newts, and moon snails), it is actually produced by certain infecting or symbiotic bacteria like Pseudoalteromonas, Pseudomonas, and Vibrio as well as other species found in animals. Parameters such as FT-–IR and Raman vibrational wavelengths and intensities for single crystal Tetrodotoxin (TTX) 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 bounds for adjacent molecules. The current study aimed to investigate the possibility of simulating the empirical values. Analysis of vibrational spectrum of Tetrodotoxin (TTX) is performed based on theoretical simulation and FT–IR empirical spectrum and Raman empirical spectrum using density functional theory in levels of F/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.

Corresponding author: Alireza Heidari, Faculty of Chemistry, California South University, 14731, Comet St. Irvine, CA 92604, American International Standards Institute, Irvine, CA 3800, USA, Email: Alireza.Heidari@calsu.us

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Keywords: Vibronic Structure, Vibrational Spectra Analysis, Density Functional Theory (DFT), Tetrodotoxin (TTX), 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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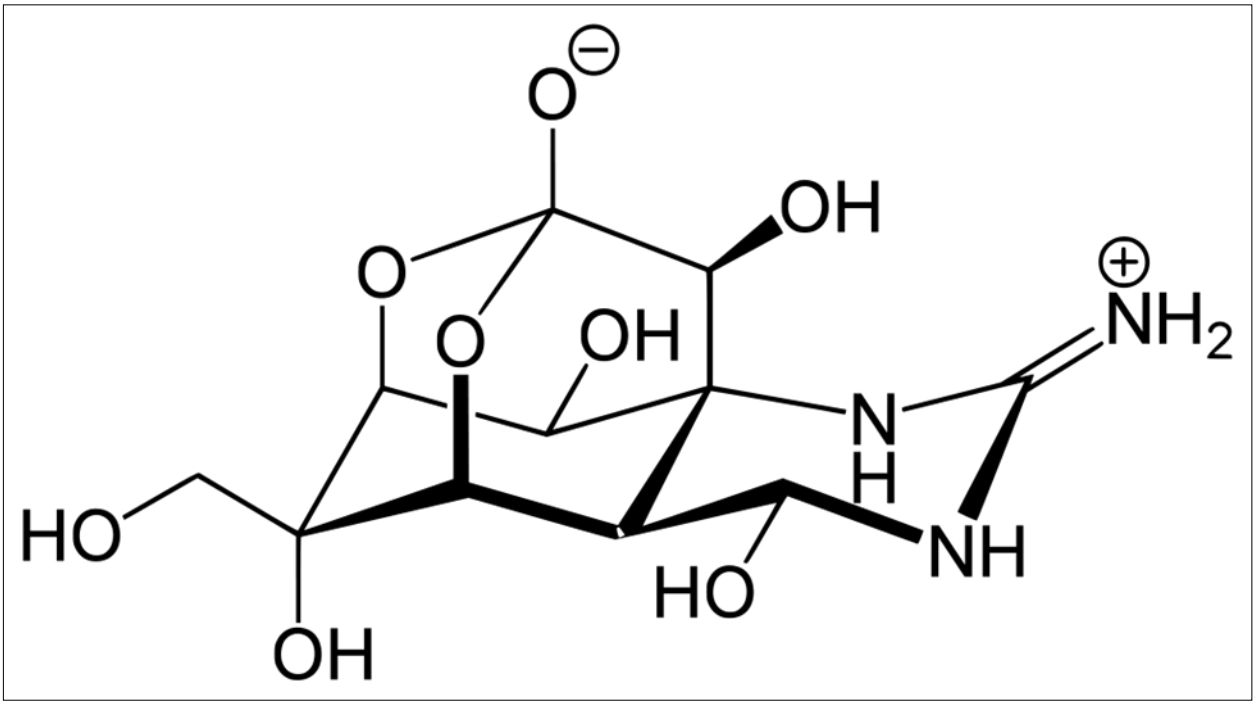

Molecular structure of Tetrodotoxin (TTX) [1–42].

Introduction

Tetrodotoxin (TTX) is a potent neurotoxin. Its name derives from Tetraodontiformes, an order that includes pufferfish, porcupinefish, ocean sunfish, and triggerfish; several of these species carry the toxin. Although tetrodotoxin was discovered in these fish and found in several other aquatic animals (e.g., in blue–ringed octopuses, rough–skinned newts, and moon snails), it is actually produced by certain infecting or symbiotic bacteria like Pseudoalteromonas, Pseudomonas, and Vibrio as well as other species found in animals. Density Functional Theory (DFT) is one of the most powerful calculation methods for electronic structures [1–7]. 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 F/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 [17-47].

Tetrodotoxin (TTX) is a tropical weed belonging to Toxins family. Amongst Toxins, Tetrodotoxin (TTX) contains anti–fungal activity, acetylcholinesterase inhibitory activity, anti–oxidant activity [48–64], mast cell stabilization and membrane protection activity [65–92], anti–bacterial activity [93–127] and anti–cancer activity [128–145], anti–hyperglycemic an anti–hyperlipidemic effects [146–156] and anti–arthritic activity, immunomodulatory activity [157–173] and anti–diabetic activity [174–188]. Our earlier report stated that the Tetrodotoxin (TTX) has high anti–oxidant activity [189–201]. The observations show that Tetrodotoxin (TTX) can be used for pharmaceutical applications. In this view, Tetrodotoxin (TTX) was taken and examined for its phytochemical and active principles in vitro anti–oxidant models and in silico approach for anti–histamine activity. Free radicals are atoms with unpaired electrons which can cause various diseases. Intake of vitamin E can reduce the problems associated with free radicals in the body [202–210]. The unpaired electrons of free–radical accumulation cause oxidative stress in the body. Oxidative stress causes cell damage leading to various health issues such as chronic disease,

cancer, autoimmune disorders, aging, cataract, rheumatoid arthritis, cardiovascular diseases, neurodegenerative diseases, respiratory disorders [211–219] and also the induced oxidative stress causes bronchial contraction by the release of cyclooxygenase and lipooxygenase in the airway that leads to bronchial asthma in human [220–226]. Asthma is a chronic inflammatory lung disease that happens due to the respiratory infection triggered by the inhalation of allergens like tobacco smoke, air pollutants, genetic and environment factors [227–233] which leads to the release of histamine and leukotrienes from the mast cell in the lung. The high release of histamine due to allergic reactions is regulated by histamine H1 receptor [234–326]. Histamine affects the immune response and related functions in human through H1, H2, H3 and H4 receptors activation with their intracellular signals [237–240]. The present research work demonstrates the chemotaxonomy of such valuable plant, from the genus of Tetrodotoxin (TTX). In addition, pharmaceutical applications such as in vitro anti–oxidant and in silico anti–histamine activity of their active principles as natural remedy was examined.

Details of Calculations

All calculations of molecular orbital in the base of ab are performed by Gaussian 09. In calculation process, the structure of Tetrodotoxin (TTX) molecule (Figure 1) is optimized and FT–IR and Raman wavenumbers are calculated using F/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 is

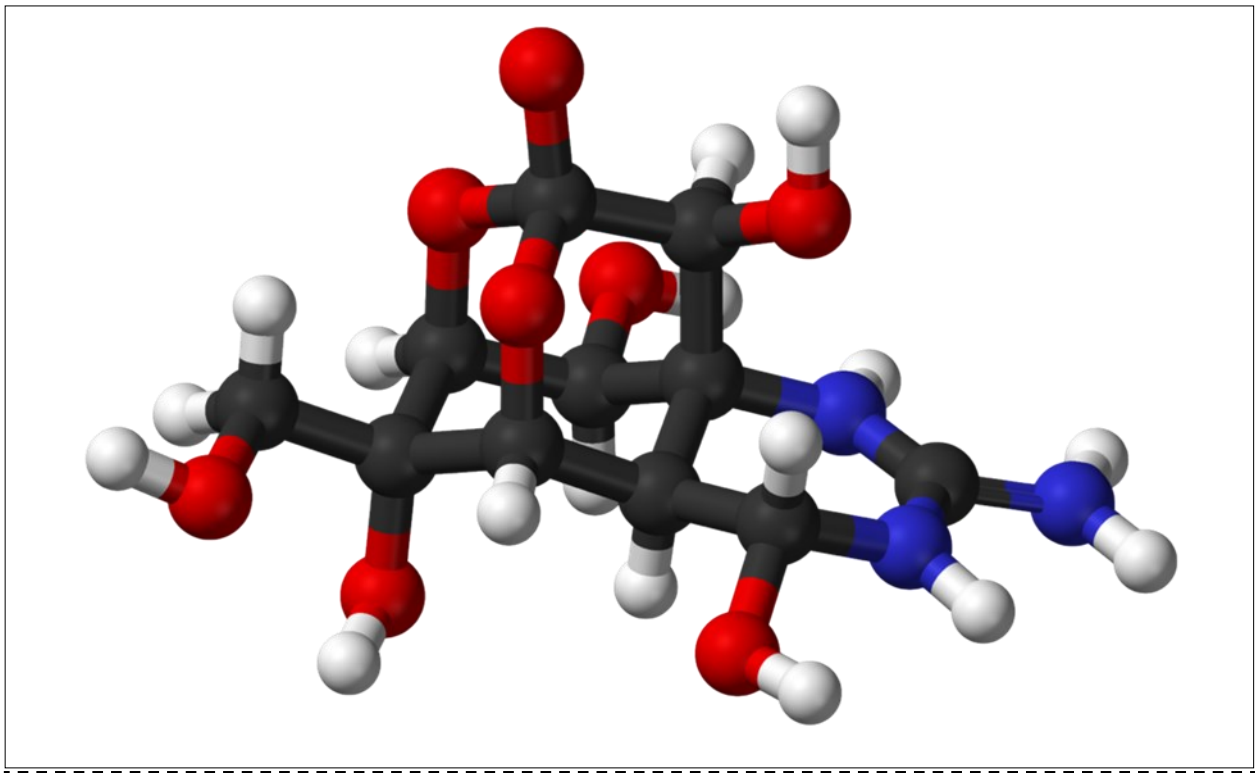

Figure 1. Section of the Tetrodotoxin (TTX) [43–93].

performed by F/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 454 (K).

Vibration Analysis

Analysis of vibrational spectrum of Tetrodotoxin (TTX) is performed based on theoretical simulation and FT–IR empirical spectrum and Raman empirical spectrum using density functional theory in levels of F/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 3255–3505 cm^{-1}. Weak Raman bands are at 3244 cm^{-1} and 3257 cm^{-1}. C–C stretching mode is a strong Raman mode at 1199 cm^{-1}. Raman weak band is seen at 1673 cm^{-1}, too. Bending mode of C–H is emerged as a weak mode at 1453 cm^{-1} and 1252 cm^{-1} and a strong band at 1346 cm^{-1} in Raman spectrum. Raman is considerably active in the range of 1255–1505 cm^{-1} which 1248 cm^{-1} indicates this issue.

C–H skew–symmetric stretching mode of methylene group is expected at 3185 cm^{-1} and its symmetric mode is expected at 3054 cm^{-1}. Skew–symmetric stretching mode of CH_2 in Tetrodotoxin (TTX) has a mode in mid–range of Raman spectrum at 3155–3275 cm^{-1}. When this mode is symmetric, it is at 3145 cm^{-1} and is sharp. The calculated wavenumbers of higher modes are at 3108 cm^{-1} and 3148 cm^{-1} for symmetric and skew–symmetric stretching mode of methylene, respectively.

Scissoring vibrations of CH_2 are usually seen at the range of 1582–1636 cm^{-1} which often includes mid–range bands. Weak bands at 1595 cm^{-1} are scissoring modes of CH_2 in Raman spectrum. Moving vibrations of methylene are usually seen at 1524 cm^{-1}. For the investigated chemical in the current study, these vibrations are at 1394 cm^{-1} were calculated using density functional theory. Twisting and rocking vibrations of CH_2 are seen in Raman spectrum at 970 cm^{-1} and 1244 cm^{-1}, respectively, which are in good accordance with the results at 954 cm^{-1} and 1229 cm^{-1}, respectively.

In a non–ionized carboxyl group (COOH), stretching vibrations of carbonyl [C=O] are mainly observed at the range of 1895–1943 cm^{-1}. If dimer is considered as an intact constituent, two stretching vibrations of carbonyl for symmetric stretching are at 1785–1840 cm^{-1} in Raman spectrum. In the current paper, stretching vibration of carbonyl mode is at 1852 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 1424 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 1084 cm^{-1} of Raman spectrum. The stretching mode of C–O (H) emerges as a mid–band of Raman spectrum at 1302 cm^{-1}.

Lattice vibrations are usually seen at the range of 0–650 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 243 cm^{-1}, 348 cm^{-1} and 407 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 F/6–31G*, HF/6–31++G**, MP2/6–31G, MP2/6–31++G**, BLYP/6–31G, BLYP/6–31++G**,

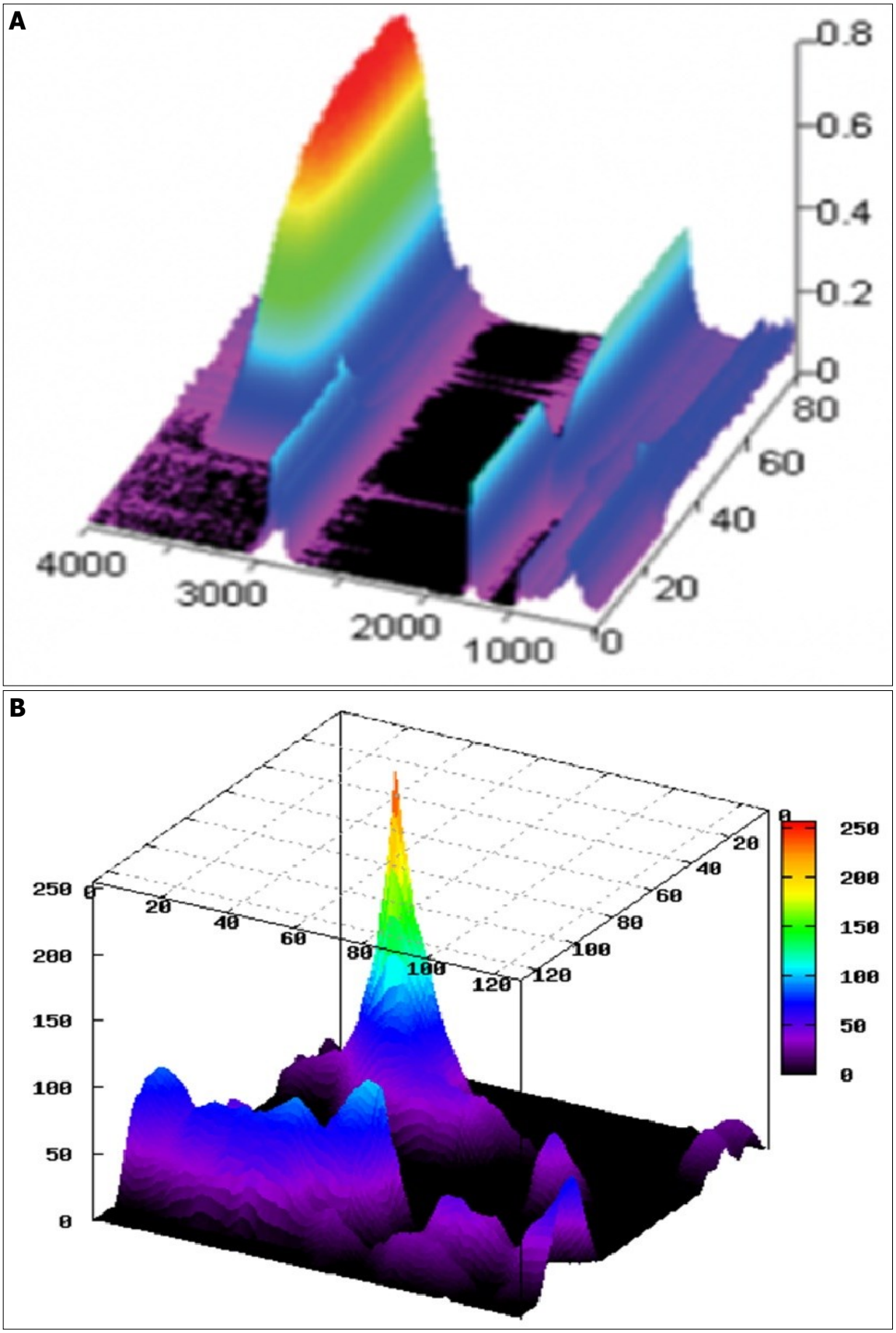

Figure 2. 3D Simulation of (a) FT–IR spectrum and (b) Raman spectrum of Tetrodotoxin (TTX).

B3LYP/6–31G and B3LYP6–31–HEG** levels were used to obtain vibrational wavenumbers and intensities in single crystal of Tetrodotoxin (TTX). 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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