A Computational Study for Optimizing a Synthetic Pathway to a Difluorinated Gingerol Compound

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Abstract

[6]-Gingerol is an abundant compound that is isolated from the roots of ginger and contains physiological and medicinal properties. [6]-Gingerol offers antiemetic, anti-inflammatory, antioxidant, and anticancer activity. However, in the body, [6]-gingerol is metabolized into [6]-shogaol during a rapid dehydration reaction by exposure to heat and acidic conditions in the body, specifically the gastrointestinal tract, and this reaction rapidly degrades the medicinal value of the natural compound. In contrast, a difluorinated gingerol compound would have, it is believed, greater stability and therefore greater therapeutic value. To optimize a synthetic pathway to produce the end product 2,2-difluorogingerol, *ab initio* molecular optimizations and calculations of vibrational frequencies and energy values were performed using the Gaussian 03 software. These calculations involve computational methods to compare differences in energies between the starting reactants vanillylacetone **1** and benzylacetone **3,** and reactant enolate intermediaries **2** and **4** (see Figure 3), as well as several other starting reactants. It is hypothesized that the presence of the methoxy (-OCH3) and hydroxyl (-OH) aromatic groups contribute to the activation of the aromatic benzene ring in vanillylacetone **1**. This suggests that vanillylacetone **1** is stable in contrast to benzylacetone **3**. This research builds on a route, previously reported by this researcher, by which vanillylacetone **1** reacts with base lithium bis(trimethylsilyl)amide (LiHMDS) to deprotonate the α-proton and generate reactive enolate. In contrast, when benzylacetone **3** reacts with base LiHDMS to generate enolate intermediate **4**, the process is very reactive. The energy values between the starting materials and the reactive enolate intermediates (see Figure 3) suggest an optimal synthetic pathway for benzylacetone **3** but not vanillylacetone **1**.

Introduction

In the last few decades, herbal resources have been identified to possess various medicinal properties and physiological effects in the body. Ginger, a potent herb, is believed to have first been harvested by the Chinese and the Indians, and is widely known for its medicinal uses. Over the years, researchers isolated ginger into its bioactive components. Among the bioactive compounds discovered were [6]-gingerol ((*5S*)-5-

hydroxy-1-(4-hydroxy-3-methoxyphenyl) decan-3-one), which has been found to exhibit pharmacological and physiological effects in the body.[1] [6]-Gingerol (Figures 1 and 2) is the most abundant bioactive component from ginger and is isolated from its roots. It is a widely recognized antiemetic for the treatment of nausea, vomiting, and the adverse side effects of chemotherapy. It has anti-inflammatory, anti-tumor, and anesthetic activities. It can also be used to treat various anxiety disorders, cardiovascular diseases, and neurodegenerative disorders like Alzheimer`s and Parkinson`s.

There is only one problem with [6]-gingerol: it is very unstable. When it is metabolized, [6]-gingerol undergoes a dehydration reaction by exposure to high temperatures and highly acidic conditions in the gastrointestinal tract. [6]-Gingerol is metabolized into [6]-shogaol ((*E*)-1-(4-hydroxy-3-methoxyphenyl) dec-4-en-3-one) by a rapid dehydration reaction due to the presence of a *β*-keto hydroxyl group (shown in Figure 1).[2] A dehydration reaction is a type of *β*-elimination reaction in organic chemistry resulting in the loss of water.

The stability of [6]-gingerol and [6]-shogaol in aqueous solutions shows that [6]-gingerol degrades to form [6]-shogaol reversibly at 100 ◦C and 1.0 pH within 2 hours.[2] The *β*-keto hydroxyl group makes [6]-gingerol susceptible to a dehydration reaction resulting in the removal of the hydroxyl (–OH) group and a hydrogen ion (H^+) to form a double bond (Figure 1).

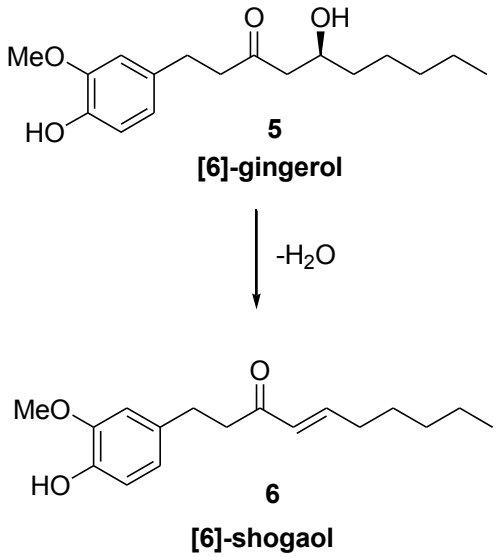


Figure 1. Dehydration from [6]-gingerol to [6]-shogaol.[2]

A difluorinated gingerol compound, it is thought, will possess greater thermal and chemical stability, and therefore greater medicinal value than [6]-gingerol. The computational methods employed in this study point to a synthetic pathway that minimizes the energies produced from steric and bulky interactions and provides an

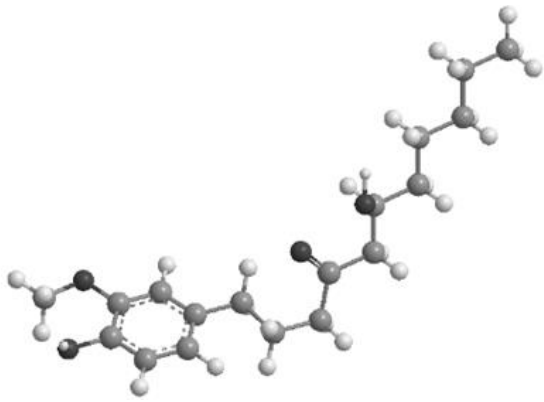


Figure 2. Three-dimensional representation of [6]-gingerol. (Structure produced from Chem3D Ultra 8.0)

energy-favorable process to produce an appropriate di-fluorinated compound: within the limits of this study, the starting reactant benzylacetone yielded efficient synthesis of difluorinated product while vanillylacetone was singularly inefficient. This research supports the claim that vanillylacetone **1** is too stable and nonreactive in contrast to benzylacetone **3,** the more reactive starting material (Figure 3). Vanillylacetone **1** did not deprotonate to yield intermediate **2** and [6]-2,2-difluorogingerol, the compound of medicinal interest.

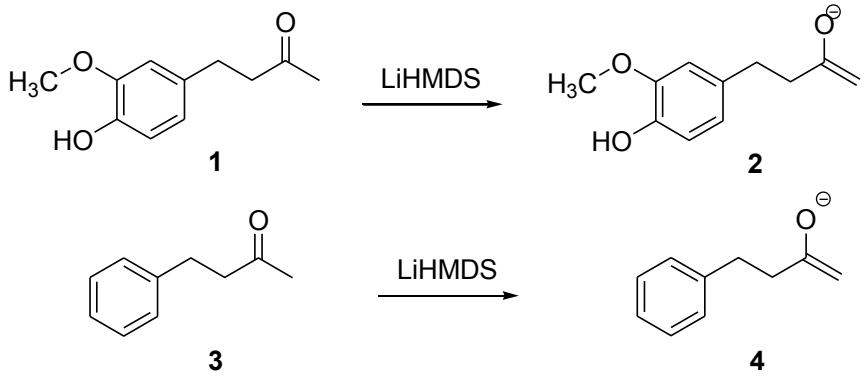


Figure 3. Formation of reactive enolates. where the starting reactants are vanillylacetone **1** and benzylacetone **3**. (Compounds produced from ChemDraw Ultra 8.0)

Hypothesis and Purpose of Research

The goal of the research is to ascertain an optimal pathway to incorporate two fluorine molecules to [6]-gingerol (Figures 1 and 2) in order to enhance thermal and chemical stability, and thus improve the therapeutic value of the modified gingerol compound.

A previous multi-step synthesis to achieve 2,2-difluorogingerol was attempted; however, the desired end product was not attained. The purpose of this computational research is to determine the causes and errors associated with previously reported synthetic routes. The hypothesis for this experiment is that, in the second step of the organic synthesis, the presence of the phenylhydroxy group (-OH group) and phenylmethoxy group (-OCH3) in vanillylacetone **1** (Figure 3) is what accounts for the low percent yield of the desired difluorogingerol end product. It is suspected that the starting material vanillylacetone **1** is too stable and not very reactive.

To ascertain a superior method of synthesis, a computer-based molecular optimization is undertaken to calculate the B3LYP energy differences using the standard *ab initio* Hartree-Fock method to distinguish between different reactants, reaction intermediates, and specific molecular conformations. The same calculations are performed on benzylacetone **3** and intermediates **2** and **4** in Figure 3, as well as on compounds depicted in Figure 4. This study will also yield the enthalpic (∠H), entropic (∠S), and Gibbs free energy (∠G) values, as well as the partial charge values, dipole moments, rotational and vibrational frequencies, and activation energies for various starting reactants.

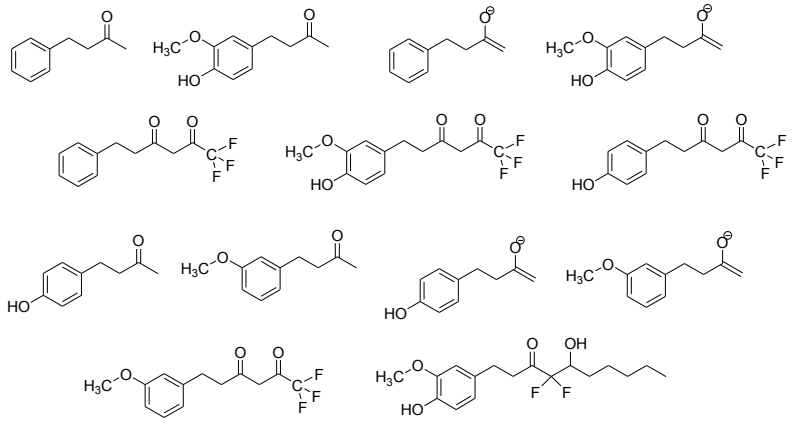


Figure 4. Library of compounds that undergo *ab initio* Hartree Fock calculations. (Structures produced from ChemDraw Ultra 8.0)

This study does not consider acidity of protons in the molecule, especially the phenolic hydrogen position ($pK_a \cong 10$). The paper is structured to help the reader understand the complex calculations associated with the research. The research does not involve biochemical analysis or compound-substrate binding studies, but instead encompasses computational and organic synthetic formulation only.

Methodology

This inquiry did not involve a synthesis of a difluorinated gingerol compound, but rather a computational study for different starting reactants and reactive intermediates as seen in in Figures 3 and 4. The materials used in this experiment were computational and drawing software. The university`s pre-installed Gaussian 03 software, the ChemSketch freeware, ChemDraw Ultra 8.0, and Chem3D Ultra 8.0 software were also used. A wide variety of compounds were studied in order to undergo molecular optimization calculations with the Gaussian 03 software.[3] Selected compounds in Figure 4 were analyzed in this research. The compounds of major focus in this study are vanillylacetone **1**, benzylacetone **3**, and reactive enolates **2** and **4** (Figure 3). Graphical representations of energy transitions were constructed for stability analysis. The calculations were logged in the university`s WebMO graphical user interface service. An optimized synthetic approach will be proposed based on the results of the calculations.

Effect of Fluorine and Its Reactivity

Fluorine exhibits many physiochemical properties that can enhance a compound`s reactivity in *in vivo* biological systems. The unique physical properties of fluorinated compounds derive, in part, from the extreme electronegativity of fluorine. The carbon-fluorine bond (C-F) is a polar bond in organic chemistry with a relatively large dipole moment compared to that of a carbon-hydrogen bond (C-H). A C-F bond is polarized in the opposite direction to a C-H bond, and is more stable (by about 14 kcal/mol) and polarizable than a C-H bond.[4] The C-F bond (1.4 Å) is significantly longer than a C-H bond (1.0 Å). Nevertheless, fluorine can frequently be substituted for hydrogen in very small nonpolar molecules, with minimal impact on their binding to proteins and enzyme complexes.[4] Fluorinated R-groups in specific hydrophobic and basic amino acid sequences of proteins exhibit a hydrophobic effect in water and have been shown to exhibit chemical and thermal stabilization in secondary and tertiary protein structures due to hydrophobic interactions.[4] Fluorine has a binding affinity with lysine, arginine, histidine, valine, leucine, and isoleucine. Such fluorinated protein analogues exhibit increased chemical and thermal stability towards the protein unfolding by chemical denaturants, solvents, heat, and degradation by protease or ubiquitin mediated complexes.[4] Overall, fluorinated compounds display an enhancement of thermal stability (C-F ~ 107 Kcal/mol), an increase of lipophilicity and hydrophobicity, increasing bioavailability, and the capability of mimicking enzyme substrate complexes.[5] Fluorinated compounds dissolve in highly acidic conditions, such as the gastrointestinal tract, and exert their physiochemical properties for their desired mechanisms of action.

Quantum Computational Chemistry

Computational chemistry has become a useful way to investigate materials and compounds that are too difficult to find or too expensive to purchase. This helps researchers make predictions before running the actual experiments so they can be better prepared for making observations. Computational chemistry and its applications are based on the Born-Oppenheimer approximations to function in the simplification of the

Schrodinger equation. The Schrodinger equation is a partial differential equation that describes how the quantum state of a quantum system changes with respect to time.[6] Therefore, the Schrodinger equation is time-dependent. The equation is not a simple algebraic equation, but a linear partial differential equation that describes a system`s electronic wave functions and configurations. The equation describes a single particle of mass moving with energy in one dimension. The particle does not have a precise trajectory; instead there is only a probability that it may be found at a specific location at any instant.[6] Erwin Schrodinger`s quantum mechanical systems state that the laws of motion and the quantum conditions are deduced from the Hamiltonian operator ($\hat{H}$).[6] The Hamiltonian operator corresponds to the total energy of the system considering the sum of the kinetic and potential energies of the system.

A definite localization of the electric charge in space and time can be associated with the wave-system and can account for electrodynamic principles for frequency, intensity, and polarizations of emitted light.[6] The equation and its adjusted approximations by Hartree-Fock theory measure the probability distributions of electrons in space. However, there is a level of uncertainty to these methods and theories. First, in classical Newtonian mechanics, a particle has an exact position and an exact momentum. In quantum mechanics, Werner Heisenberg states that a particle may have a definite position, but a definite velocity or momentum cannot be known to be precise.[7] The Schrodinger equation cannot supply a specific measurement of the wave function even if the wave function is known exactly. Only predictions and approximations can be made.

Other approximation methods are made by Born and Oppenheimer to simplify the Schrodinger equation. The Born-Oppenheimer approximation simplifies the Schrodinger equation by noting that the position of the atomic nucleus remains relatively constant due to the nucleus having a mass larger than that of the oscillating electrons. The Born-Oppenheimer equations neglect the velocities of nuclei, and the nucleus is assumed to be stationary while electrons move around it. The motion of the nuclei and the electrons can be separated, and the electronic and nuclear configurations can be solved with independent wave functions.[8] Quantum mechanical methods are suitable for calculating molecular orbital energies and coefficients, heats of formation of specific conformations, partial atomic charges, electrostatic potentials, dipole moments, transition-state geometries and energies, and bond dissociation energies.

Ab Initio Method of Calculation

Ab initio methods involve performing molecular dynamics calculations on a system of nuclei without prior knowledge of the quantum mechanical potential energy surface.[9] These calculations account for the potential energy difference of electrons in the excited states altogether. The minimum basis set for atomic orbitals follows the STO-3G basis set for Gaussian integrals (Figure 5). STO-3G stands for Slater-type orbital (STO) and measures the three Gaussian orbitals (1s, 2s, and 2p). Slater-type Orbitals are accurate; however, it requires longer to calculate integrals using STO basis sets. Instead, Gaussian integrals are computed to approximate the STO values by STO-nG basis sets. Therefore, quantum chemists and physicists use a linear combination of enough Gaussian-type Orbitals (GTOs) to mimic a STO basis set (Figure 6). In quantum

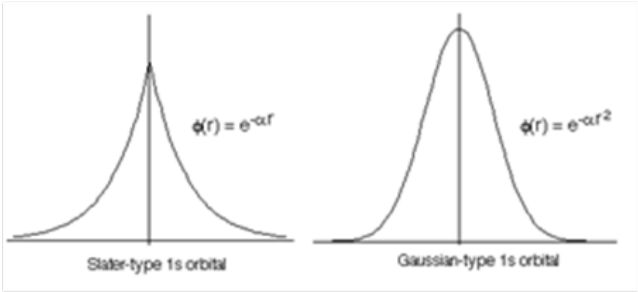


Figure 5. STO-1s vs. GTO-1s basis functions.[11]

chemistry, the basis set usually refers to the set of one-particle functions used to build molecular orbitals from atomic orbitals, where an orbital is a one-electron function. The primitive Gaussian functions (ϕ_i) are represented by the core and valence electrons surrounding the central atom. This basis set works for large systems in *ab initio* methods used primarily for geometric optimizations. This method, observed in Figure 6, involves

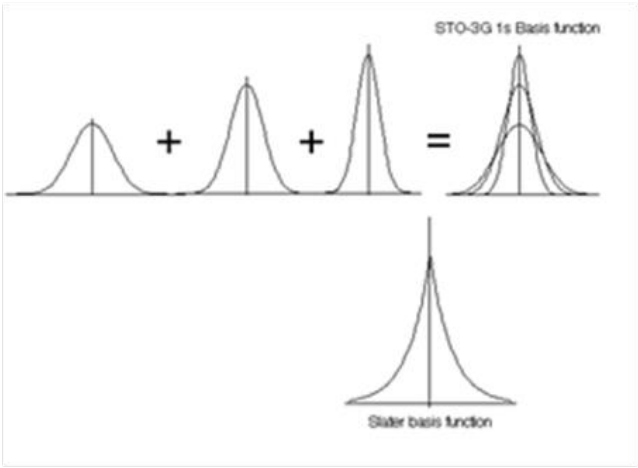


Figure 6. STO-3G basis function.[11]

approximating the Gaussian integrals of an STO-3G basis set with fitting functions in order to decrease the computational costs of evaluating the energy and forces during molecular simulations.[9] This method is an *ab initio* one and will give results with accuracy similar to those obtained from an STO-3G calculation. It can easily be extended to larger basis set calculations like 6-311G (p,d,f) as well.[9] The *ab initio* calculations performed follow the general Hartree-Fock theory in conjunction with a standard Gaussian basis set (STO-3G). The Hartree-Fock theory states that an electron's motion is a single-particle function or Slater orbital that does not depend on the instantaneous motions of the other electrons.[10] The Hartree-Fock (HF) theory essentially is another approximation to the Schrodinger equation that expresses the total wave function of a system as a product of one electron occupied in atomic orbitals. The motions of all other electrons are taken as an average distribution only.

Previous Synthesis and Research

A previous study of [6]-2,2-difluorogingerol used the trifluoroacetate release method to generate highly reactive intermediates, but the study did not yield the desired difluorinated gingerol intermediate **2** in Figures 3 and 7.[12] The trifluoracetate release strategy was developed at Purdue University and is proven to generate reactive intermediates for a variety of compounds.[12] The researchers describe the application of the trifluoroacetate-release strategy to cleave a carbon-carbon bond and generate α, α-difluoroenolates. This method uses exceedingly mild reaction conditions, is not limited to a small group of fluorinated building blocks, and is compatible with many substrates.[12] The problem with the previous synthesis was the first step of the reaction. The previous synthesis by method of trifluoroacetate release to generate [6]-2,2-difluorogingerol by carbon-carbon selective cleavage strategy through a sequential aldol condensation reaction did not yield as anticipated. Molecular optimization of the reaction conditions and intermediates can be made to provide molecular and quantum-based calculations for better geometric optimizations and compound stability. Figure 7 illustrates the four step synthesis performed in previous works. The problem encountered occurred in the first step of the reaction involving vanillylacetone **1** and base LiHDMS to produce intermediate **7**.

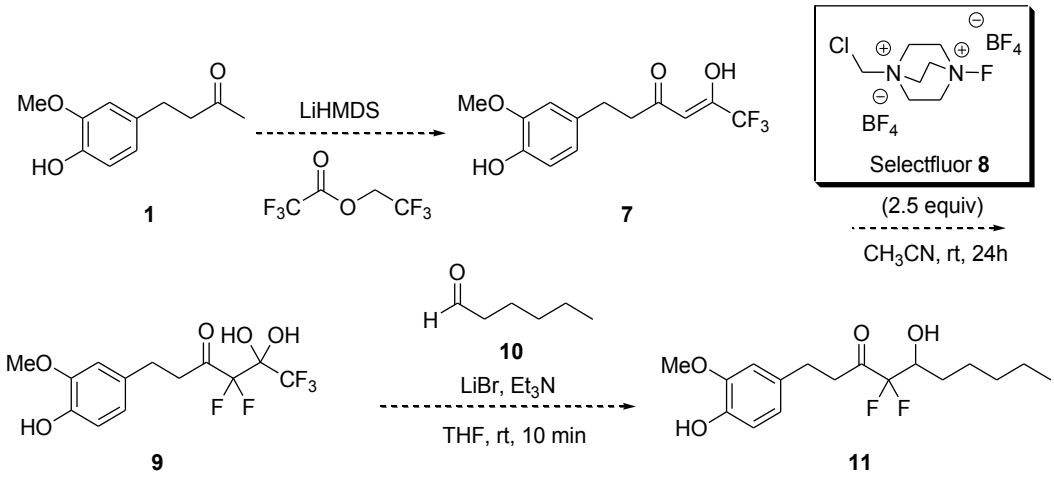


Figure 7. Attempted synthesis of [6]-2,2-difluorogingerol through trifluororelease strategy.[11] (Structures produced from ChemDraw Ultra 8.0 and ChemSketch Freeware)

Results and Discussion

The two first-step reactions in Figures 8 and 9, in which a base such as LiHDMS (Figure 3) requires energy to deprotonate a hydrogen atom at the alpha carbon, are nonspontaneous processes as indicated by Hess`s Law. The reactions are endergonic/endothermic, indicating that energy is absorbed or required for deprotonation of the hydrogen atom to occur. The H^+ ion has no energy (0 Hartree) since there are no electron interactions at the electron-deficient hydrogen atom. The reaction in Figure 8 has a net Gibbs free energy of +0.815999 Hartree (~ +2,142.41 kJ/mol) and an activation energy of +0.81426 Hartree (~ +2137.84 kJ/mol). The

reaction in Figure 9 has a net Gibbs free energy value of +0.739635 Hartree (~ +1941.9 kJ/mol) and an activation energy of +0.741383 Hartree (~ +1946.5 kJ/mol). Therefore, the reaction in Figure 8 has a higher activation energy than the reaction in Figure 9 and is still a nonspontaneous process. The selection of a very strong base is required for deprotonation at the alpha carbon position.

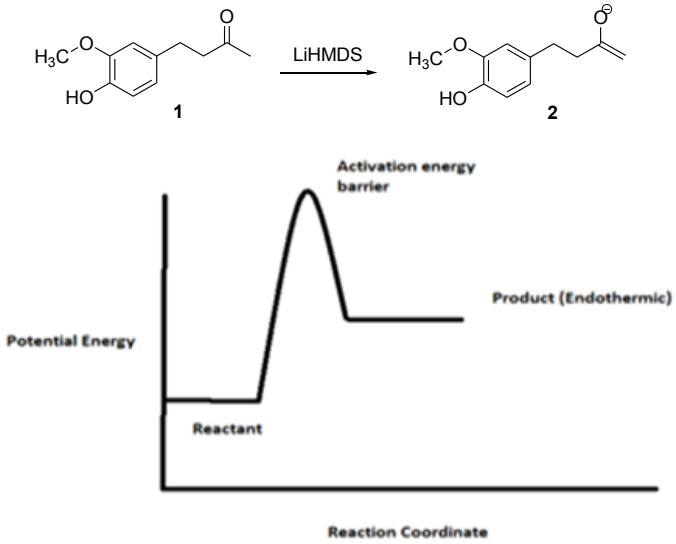


Figure 8. Energy diagram for vanillylacetone **1** and intermediate **2** under reaction with base LiHDMS. Activation energy: +0.81426 Hartree (+2,137.84 kJ/mol), Gibbs Free energy: +2,142.41kJ/mol.

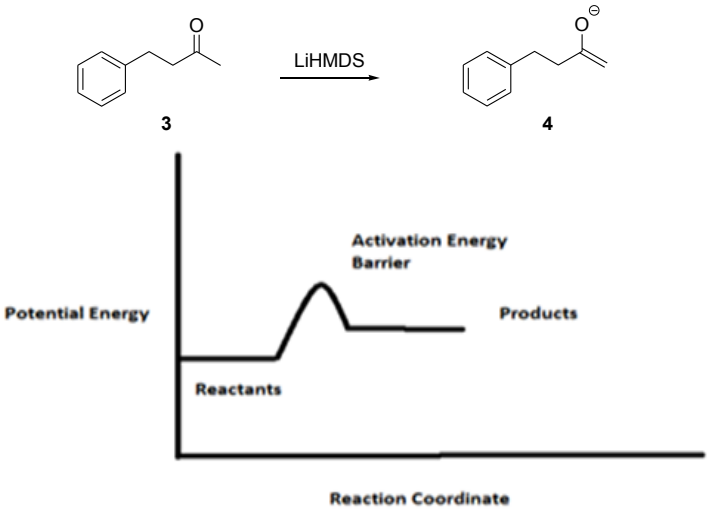


Figure 9. Energy diagram for benzylacetone **3** and intermediate **4**, respectively. Activation energy: +0.741383 Hartree (+1946.5 kJ/mol), Gibbs Free energy: +1941.9 kJ/mol.

The reaction in Figure 8 may have a large activation barrier; however, it is the preferred reaction since the desired difluorinated gingerol compound requires vanillylacetone as the starting material. With vanillylacetone as the starting reactant, the end product 2,2-difluorogingerol contains the two phenyl R groups required for the desired course of action.

In terms of calculating energy values for small molecules, the greater the number of atoms, the lower the energy values and the greater the stability. Molecules with a lower number of atoms correspond to higher energy values and greater reactivity as evident in Figures 10 and 11. The basis set for calculations on the Gaussian 03 software followed a 6-311+G(2p) basis set for vanillylacetone **1** and benzylacetone **3** (Figures 10 and 11).[3] The basis set for intermediates **2** and **4** followed a 6-31G(d) basis set (Figures 10 and 11).[3] The difference in basis sets for each compound lies in the outer electron calculation. Vanillyacetone **1** and intermediate **2** are very stable compounds and not as reactive based on the calculated data. Benzylacetone **3** and intermediate **4** are very reactive compounds and more likely than not can undergo the reaction with the conditions outlined in Figure 7. It is reported that benzylacetone **3** underwent mild trifluoroacetate release via carbon-carbon selective cleavage to yield a difluorinated end product.[12] Vanillylacetone **1** (Figure 3), however, did not undergo the reactions to yield difluorinated enolates or products. The chemical structure and identity of [6]-gingerol in Figures 1 and 2 encompass the two phenyl groups attached to the compound that are important for [6]-gingerol`s medicinal activity. The presence of the two phenyl groups (-OCH_3 and –OH) on [6]-gingerol, observed in Figures 1 and 2, is crucial for the activation of the aromatic benzene ring in [6]-gingerol and the compound's medicinal activity. Both –OCH_3 and –OH groups are electron-donating substituted R groups that function in the activation of the aromatic benzene ring.

The problem in the synthesis in previous works was that the initial step in the formation of the enolate did not work. The base LiHDMS is a weak, sterically hindered base that did not deprotonate the hydrogen atom at the a_2 carbon position. In organic chemistry, generation of enolates occurs when a strong or relatively strong nucleophilic base deprotonates a hydrogen atom at the α carbon position. The *a* carbon is adjacent to the carbonyl (C=O) group. There are two carbon atoms adjacent to the carbonyl group, which poses a problem as illustrated in Figure 12. A stronger and less bulky base must be used to deprotonate the hydrogen atom at the a_2 position so Zaitsev`s rule cannot be satisfied. Zaitsev`s rule states or predicts that in an elimination reaction, the most stable alkene is the most substituted carbon atom. This reaction with base is not a β-elimination reaction, but a reaction involving the formation of enolates. Instead, for a future synthesis, nBuLi base will be added to vanillylacetone and will deprotonate at the a_2 position. The reaction must occur at low temperature as deprotonation will occur faster than at high temperatures. The base nBuLi illustrated in Figure 13, is a very strong, non-sterically hindered base that will deprotonate and result in the formation of a kinetically preferred reactive enolate shown in Figure 12.

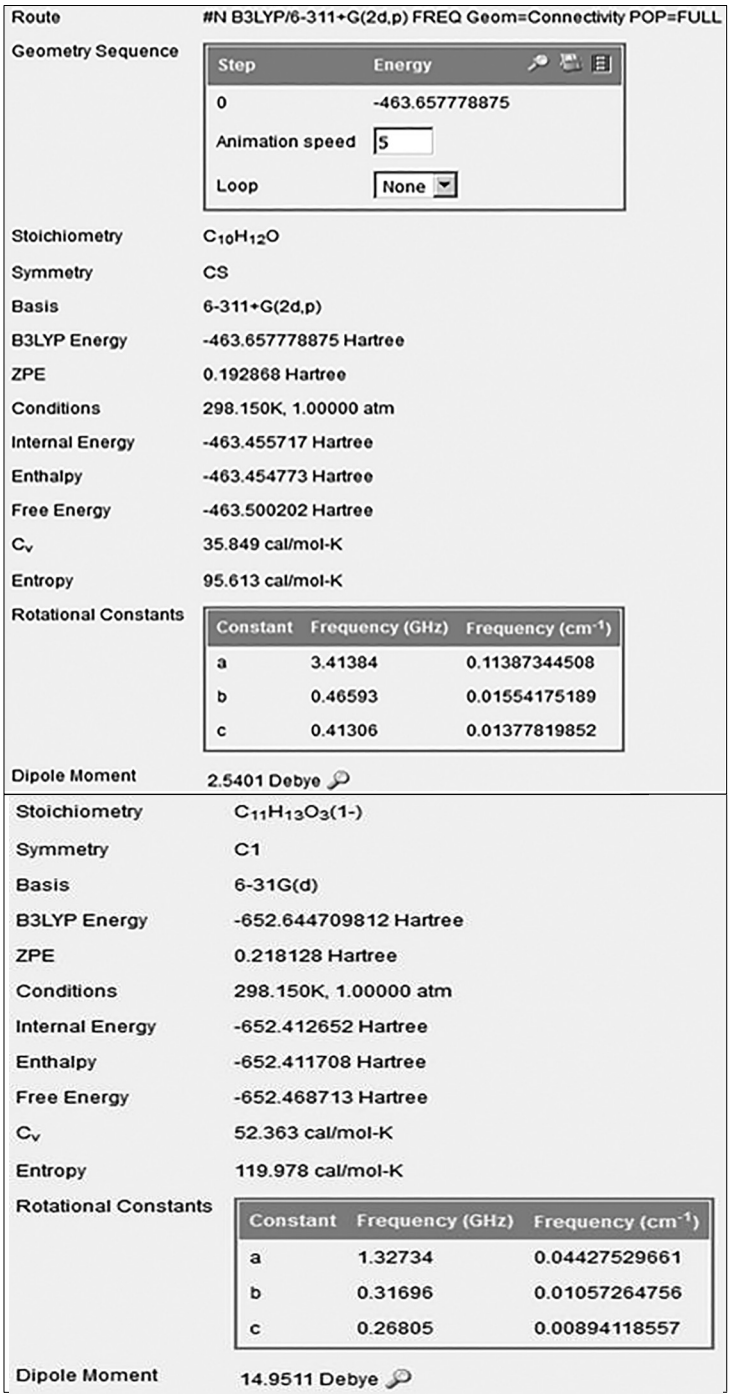
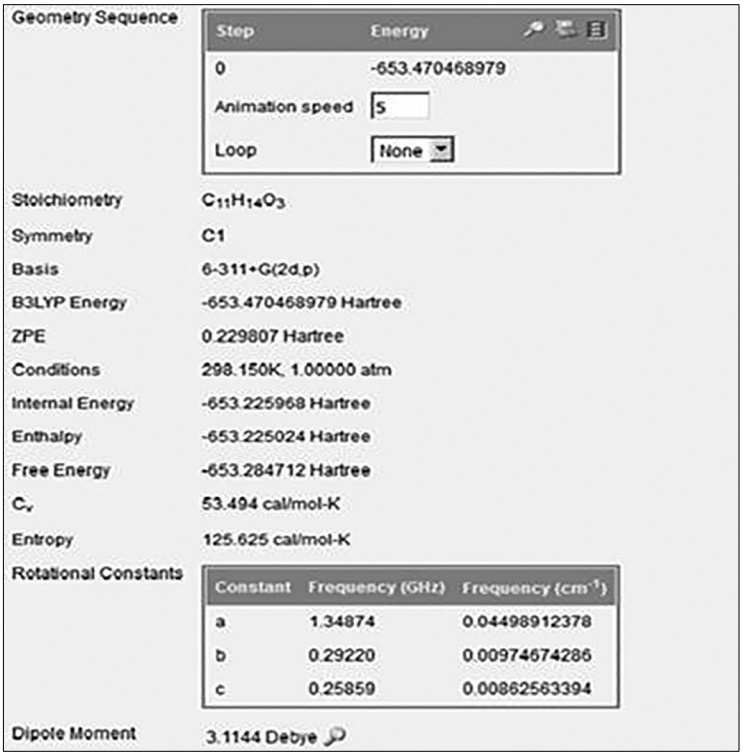


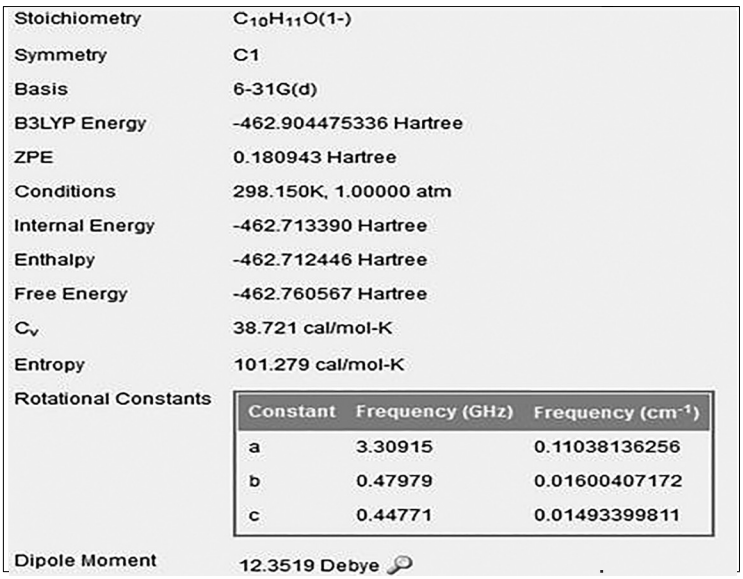
Figure 10. *Ab initio* calculations for vanillylacetone **1** and intermediate **2**, respectively.[3] (Results provided by WebMO services and Gaussian 03 software)



Geometry Sequence		
Step	Energy	
0	-653.470468979	
Animation speed	5	
Loop	None	
Stoichiometry	$C_{11}H_{14}O_3$	
Symmetry	C1	
Basis	6-311+G(2d,p)	
B3LYP Energy	-653.470468979 Hartree	
ZPE	0.229807 Hartree	
Conditions	298.150K, 1.00000 atm	
Internal Energy	-653.225968 Hartree	
Enthalpy	-653.225024 Hartree	
Free Energy	-653.284712 Hartree	
C_v	53.494 cal/mol-K	
Entropy	125.625 cal/mol-K	
Rotational Constants		

Constant	Frequency (GHz)	Frequency (cm^{-1})
a	1.34874	0.04498912378
b	0.29220	0.00974674286
c	0.25859	0.00862563394

Dipole Moment: 3.1144 Debye



Stoichiometry	$C_{10}H_{11}O(1-)$
Symmetry	C1
Basis	6-31G(d)
B3LYP Energy	-462.904475336 Hartree
ZPE	0.180943 Hartree
Conditions	298.150K, 1.00000 atm
Internal Energy	-462.713390 Hartree
Enthalpy	-462.712446 Hartree
Free Energy	-462.760567 Hartree
C_v	38.721 cal/mol-K
Entropy	101.279 cal/mol-K
Rotational Constants	

Constant	Frequency (GHz)	Frequency (cm^{-1})
a	3.30915	0.11038136256
b	0.47979	0.01600407172
c	0.44771	0.01493399811

Dipole Moment: 12.3519 Debye

Figure 11. *Ab initio* calculations for benzylacetone **3** and intermediate **4**, respectively.[3] (Results provided by WebMO services and Gaussian 03 software)

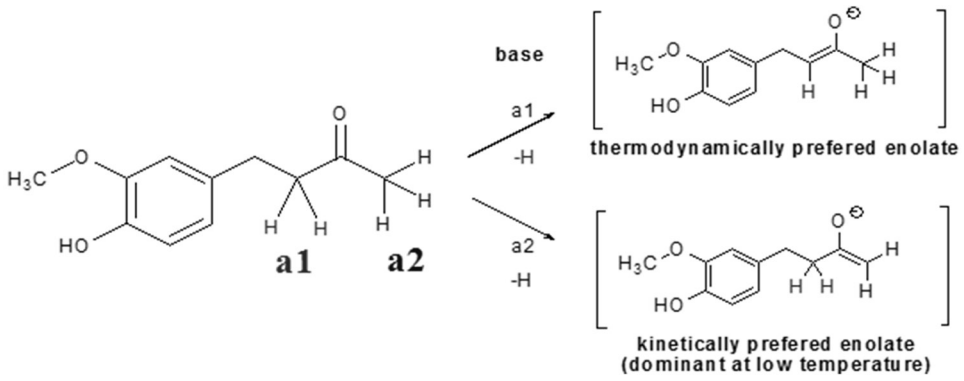


Figure 12. Deprotonation by base yields two possible temperature dependent enolate intermediates. (Structures produced by ChemDraw Ultra 8.0 software)

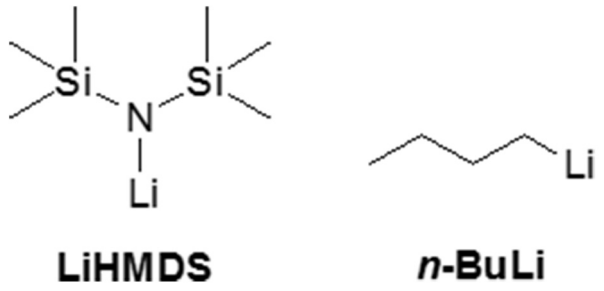


Figure 13. Structures of LiHMDS and n-BuLi nucleophilic bases, respectively. (Structures produced by ChemDraw Ultra 8.0 software)

Conclusions

This computational study for optimizing the multi-step synthesis of 2,2-difluorogingerol incorporated approximations in quantum mechanics to determine the energy values for various compounds and intermediates in Figures 3 and 4. The general Hartree-Fock *ab initio* method performed calculations on 6-311G(p,d) basis sets to approximate Slater-type Orbital (STO) functions by fitting Gaussian functions to decrease the computational costs.[9] While the computational calculations extend to all compounds in Figure 4, this study focuses on the compounds in Figure 3. Vanillylacetone **1** is more stable than benzylacetone **3** (Figure 3) due to smaller (∠G) Gibbs energy value. A large activation energy barrier is observed when vanillylacetone **1** is converted to an enolate intermediate **2** upon deprotonation by base LiHMDS (Figure 8). A small activation energy barrier is observed when benzylacetone **3** is converted to

enolate intermediate **4** upon deprotonation by base LiHMDS (Figure 9). The initial hypothesis states that in the second step of the organic synthesis, the presence of the phenylhydroxy group (-OH group) and phenylmethoxy group (-OCH3) in vanillylacetone **1** (Figure 3) is what accounts for the low percent yield of the desired difluorogingerol end product when undergoing the reaction conditions in Figure 7. It is suspected that the starting material vanillylacetone **1** is too stable and not very reactive. The hypothesis is accepted and proven correct based on computational data in Figures 10 and 11. The reaction whereby benzylacetone **3** is converted to intermediate **4** under the same conditions in Figure 7 succeeded in yielding a difluorinated end product.[12] The reaction works well for benzylacetone **3** as the starting material in Figure 3 but not vanillylacetone **1**. To correct this, a stronger base such as n-BuLi (Figure 13) can be substituted for LiHMDS (Figure 13). Vanillylacetone **1** (Figure 3) is the preferred starting material as it contains electron-donating R groups that activate the aromatic benzene ring responsible for [6]-gingerol`s (Figure 2) medicinal activity. An optimization of the reaction pathway to achieve a difluorinated gingerol compound is well underway.

References

[1] Bode AM, Dong Z. The amazing and mighty ginger. Herbal Medicine: Biomolecular and Clinical Aspects 2011:2.

[2] Bhattarai S, Tran VH, Duke CC. Stability of [6]-gingerol and [6]-shogaol in simulated gastric and intestinal fluids. Journal of Pharmaceutical and Biomedical Analysis 2007, 45:648-653.

[3] Fukuda R, Hasegawa J, Ishida M, Nakajima T, Honda Y, Kitao O, Nakai H, Klene M, Li X, Knox JE, Hratchian HP, Cross JB, Bakken V, Adamo C, Jaramillo J, Gomperts R, Stratmann RE, Yazyev O, Austin AJ, Cammi R, Pomelli C, Ochterski JW, Ayala PY, Morokuma K, Voth GA, Salvador P, Dannenberg JJ, Zakrzewski VG, Dapprich S, Daniels AD Strain MC, Farkas O, Malick DK, Rabuck A, Raghavachari K, Foresman JB, Ortiz JV, Cui Q, Baboul AG, Clifford S, Cioslowski J, Stefanov BB, Liu G, Liashenko A, Piskorz P, Komaromi I, Martin RL, Fox DJ, Keith T, Al-Laham MA, Peng CY, Nanayakkara A, Challacombe M, Gill PMW, Johnson B, Chen W, Wong MW, Gonzalez C, and Pople JA. Gaussian 03, Revision C.02. Wallingford CT: Gaussian, Inc., 2004.

[4] Buer B, Marsh E. Fluorine: A new element in protein design. Protein Science 2012 21:453-462.

[5] Park BK, Kitteringham NR, O'Neill PM. Metabolism of fluorine-containing drugs. Annu Rev Pharmacol Toxicol 2001 41:443-470.

[6] Schrödinger, E. An undulatory theory of the mechanics of atoms and molecules. Physical Review 1926 28(6):1049-1070.

[7] Heisenberg, W. Uber quantentheoretische Umdeutung kinematischer und mechanischer Beziehungen. *Zeitschrift fur Physik* 1925 33:879-893.

[8] Born M, Oppenheimer R. Zur Quantentheorie der Molekeln. Ann. Phys 1927 389:453-484. doi:10.1002/andp.19273892002

[9] Hammes-Schiffer S, Anderson HC. *Ab initio* and semiempirical methods for molecular dynamics simulations based on general Hartree-Fock theory. J. Chem. Phys, Department of Chemistry, Stanford University 1993 99:523-532.

[10] Sherrill DC. An Introduction to Hartree-Fock Molecular Orbital Theory. School of Chemistry and Biochemistry, Georgia Institute of Technology 2000.
[11] Computational Quantum Chemistry. LibreTexts. University of California at Davis: https://chem.libretexts.org/Libretexts/University_of_California_Davis.html (accessed Nov 20, 2016).
[12] Han C, Kim EH, Colby DA. Cleavage of carbon-carbon bonds through mild release of tri-fluoroacetate: Generation of α, α-difluoroenolates for aldol reactions. J. Am. Chem. Soc. 2011 133:5802-5805