

COMPUTATIONAL ANALYSIS OF PROPOSED TETRAFUROMACROLIDE

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Abstract

Comparison of the free energy of a proposed tetrafuromacrolide and the known ionophore, nonactin, during the complexation of NaSCN and KSCN salts are presented. The lowest energy conformers were generated via an MMFFaq conformer search in the molecular modeling program Spartan '14. Comparison with CSD (Cambridge Structural Database) confirmed that the predicted structures of the nonactin complexes were reasonable. The program MOPAC 2012 was then utilized with PM7 calculations to determine heat of formation and entropy of the aqueous structures. Calculations were solvated with COSMO methods. Resulting thermodynamic data demonstrates that the proposed macrolide shows a distinct selectivity for potassium over sodium. A larger difference in the ΔG_{com} suggests that nonactin has greater selectivity for K^+ over Na^+ than the proposed tetrafuromacroclycle.

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Introduction

Ionophores are a class of molecules that complex ions in their polar interior cavity. The complex can then move into a nonpolar medium due to its lipophilic exterior. Nonactin is a 2,5-derivatized tetrafuromacrolide that complexes alkali metal ions as shown in Figure 1.¹ Natural and synthetic macrocycles share common characteristics in terms of binding metals, mainly the size of the cavity and the presence of heteroatoms.²

Nonactin, a known antibiotic, functions by disrupting crucial ion gradients needed for cellular functions.³ Nonactin selectively binds K^+ over Na^+ due, in part, to sodium ion's increased enthalpy of solvation.^{3,4} Previous research showed interesting findings in the comparison of a proposed 2,3-derivatized tetrafuromacrolide (Figure 2) to nonactin.⁵ CAChe, MNDO-PM5 calculations were used to compare the binding energies and conformations of the proposed tetraester macrocycle/cation complexes to known nonactin/cation complexes and found that they were very similar but showed less coordinate covalent bonds to the ligating metal than expected. This research will use computational methods that may more accurately represent the ligated metal cations under aqueous conditions and present a clearer picture as to whether or not the proposed macrocycle could be an effective ionophore.

Computational Methods

The computational chemistry software Spartan⁶, MOPAC⁷ and HyperChem⁸ were employed to determine heat of formation

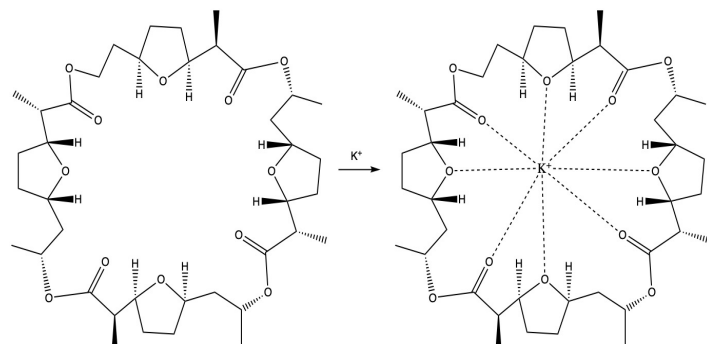


Figure 1: Nonactin can complex with potassium ions with four furan and four carbonyl oxygens.

and entropy for salts, free macrolides and complexes. Previous work found good agreement between PM6 and DFT calculations with ruthenium complexes.⁹ The semiempirical calculations were chosen because they were readily available, less time consuming, and adding in MOPAC allowed a deeper exploration of the coordinate covalent bonded complexes as compared to our previous studies. Free energy of the complexation was determined via the following calculation:

$$\Delta G_{\text{com}} = \Delta H_{\text{com}} - T\Delta S_{\text{com}}$$

The presence of alkali metals and coordinate covalent bonds in the complexes presented unique challenges for the computational program Spartan '14 which utilizes a solvation method (SM5.4 solvation) that is not parameterized for alkali metals during conformational searches. Additionally, conformer searches performed using molecular mechanics methods did not sufficiently recognize non-bonded interactions such as the coordinate covalent bonds between the metal ion and the ligand. Consequently, it was necessary to run the conformation search with constraints on the metal-ligand interactions. The presence of constraints on the molecule artificially raises the energy around those moieties, limiting the conformation search in the metal-ligand region.

Due to Spartan's limitations with handling alkali metals in molecular mechanics, the lowest energy conformer was exported to MOPAC 2012 where an additional local minima search was performed on the structure. MOPAC utilizes PM7 calculations, which have more accurate parameterization for alkali metals. MO-

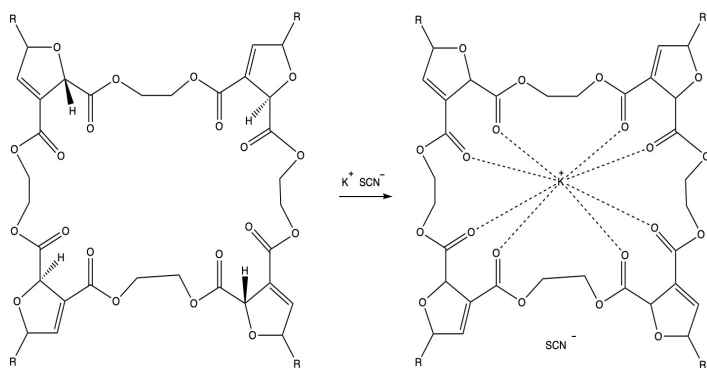


Figure 2: Proposed 2,3-derivatized tetrafuromacrolide binding with potassium ion via carbonyl oxygens.

PAC also has the ability to interpret non-bonded interactions so the constraints on the metal-ligand distances could be removed, and the metal-ligand conformation region is more fully explored.⁷ Furthermore, MOPAC uses COSMO methods for solvation, which envisions the solvent as a continuous dielectric medium and its parameterization includes alkali metal solutes.¹⁰ MOPAC needs another program to visualize the optimized structure. HyperChem 8 was chosen as a graphical user interface. JMOL was additionally utilized for visualization.

Molecules were built in HyperChem 8 as planar structures with stereochemistry then imposed on asymmetric carbons. Both nonactin and the tetrafuromacrolide followed an (*R-S-R-S*) pattern of stereochemistry around the ring. 3D structures were predicted with the invoke model builder feature. Complexes were modeled with thiocyanate salts, which were chosen because complexed nonactin species were available in the literature. The structures were imported into Spartan[®] 14 as a pdb file in order to perform a molecular mechanics conformer search. The lowest energy conformer was obtained with an equilibrium conformer calculation followed by a conformer distribution. Both conformer searches utilized Monte Carlo methods with MMFFaq calculations and SM5.4 solvation. Metal-ligand bond lengths were constrained on the complexes during the conformer search and the keyword “KEEPALL” was used to retain all plausible conformers.

The lowest energy structure was converted to a moprt file and imported into MOPAC 2012 where it was further optimized with PM7 calculations and COSMO solvation methods. Water was specified as the solvent using its dielectric constant, 78.3. Constraints on the complexes were removed for this optimization. Final structures were converted to hin files so they could be visualized in HyperChem 8. MOPAC 2012 was again utilized with PM7 calculations and COSMO solvation methods to determine heat of formation and entropy on the optimized structures. These were then used to calculate the Gibb's Free Energy change for the complexation reactions. All calculations were performed at 298 K. Conversions between file formats were carried out using the outside program Babel.¹¹

Results and Discussion

Thiocyanate salts:

Although an ionized thiocyanate salt was used as the initial structure, the structure produced by optimization in MOPAC showed a bond between the metal and sulfur atom. It is possible that this unusual structure was a by-product of COSMO's “continuous dielectric constant” approach, which does not account for solvent-solute interaction. Since thiocyanate salts are so small, the limited approach of COSMO is not sufficient to visualize them as independent ions.

Thermodynamic values were calculated for the thiocyanate salts and compared to NIST (National Institute of Standards and Technology) published NBS (National Bureau of Standards Publications) findings.¹³ The computational values did not agree with the published values for the nonactin complex (Table 1). Thus it was necessary to find alternative values. It was determined that two ΔG_{com} calculations would be run in parallel: one with thermodynamic values for MSCN (Macrocycle/ SCN^- complex)

calculated in MOPAC and one with thermodynamic values for MSCN taken from the NBS. Since NBS values represent experimentally determined data, they will necessarily represent fully ionized species and will factor in the interaction between the salts and individual water molecules.

Complexes:

The results for the structures of the optimized nonactin complexes were compared to the structures obtained from the CSD^{13a} as shown in Figure 3.

Special attention was paid to metal-ligand bond lengths as the computation programs utilized tended to have difficulty recognizing metal-ligand interactions. The structure produced by optimization in COSMO had slightly longer bond lengths than the CSD structure. Although the final optimized structures looked very plausible, they did not exactly reproduce the CSD structures. Whether this due to the differences that arise when considering two different phases (crystal vs. aqueous) or a result of inaccuracy in the calculations is difficult to determine. It is likely a mixture of both. The optimized structure of the tetrafuromacrolide complexes had slightly longer bond lengths than nonactin complexes, but they still reasonably depicted an interaction between the oxygen and metal atoms. The tetrafuromacrolide/ Na^+ complex complexed the metal ion with only carbonyl oxygens but the tetrafuromacrolide/ K^+ complex relied on ester oxygens to bind the metal ion in two instances, using carbonyl carbons for the six other ligating atoms. Unlike our previous work, these calculations displayed the correct number and location for the known coordinate covalent bonds found in the complexed nonactin structure. Unfortunately, this work shows that nonactin may be more effective in transporting the cations across the lipid membrane. The carbonyl and furan oxygens are ligated to the center cation and thus provide a lipophilic exterior for the complex, while the unbound furan oxygens of the tetrafuromacrolide would decrease this effect (Figure 3).

Table 1 – Accuracy of MOPAC thermodynamic calculations

Salt	Value	MOPAC	NBS	% Error
NaSCN	ΔH_f (kJ/mol)	-182.5	-163.7	11.52
	S (J/mol)	293.6	203.3	44.41
KSCN	ΔH_f (kJ/mol)	-67.5	-175.4	61.55
	S (J/mol)	317.8	246.9	28.73

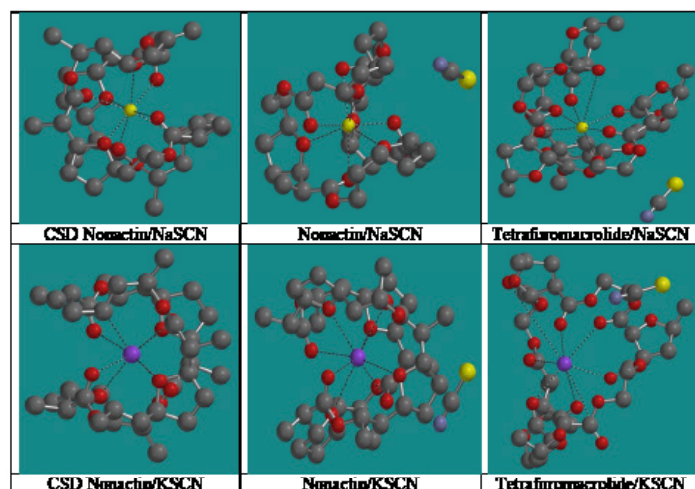


Figure 3: Structures of the nonactin and tetrafuromacrolide complexes of both NaSCN and KSCN salts produced by the modeling procedure. Structures of nonactin complexed with NaSCN and KSCN obtained from the CSD are provided for comparison.^{13b, 13c}

Regardless of whether MSCN data came from MOPAC or the NBS; ΔH_{com} , ΔS_{com} and ΔG_{com} values demonstrated that the proposed macrolide exhibited the same thermodynamic trends as nonactin (Tables 2 & 3). Calculated ΔG_{com} was more negative for the potassium complexes than the sodium complexes in the case of both nonactin and the tetrafuromacrolide, demonstrating that both macrolides display selectivity for K^+ over Na^+ (Table 2). The same trend was observed whether data for the thiocyanate salt was taken from MOPAC or from the NBS (Table 3).

Interestingly, the difference in $-\Delta G_{\text{com}}$ between KSCN and NaSCN complexes was much larger when nonactin was the ligating ring as compared to the tetrafuromacrolide complex (Table 4). This suggests that, while the proposed macrolide is selective for K^+ over Na^+ , it may not be selective to the degree that nonactin is. The difference in $-\Delta G_{\text{com}}$ was greater when thermodynamic values for MSCN were calculated by MOPAC than when they were taken from the NBS (Table 4) for both macrocycles. This discrepancy is connected with the errors observed earlier when calculating thermodynamic data with COSMO assuming poorly ionized thiocyanate salts. However, since the same overall trends were observed in both instances, the error on the thiocyanate calculations did not have an effect on the overall conclusion.

A few limitations of the computational programs impacted the accuracy of the calculated thermodynamic results. In particular, COSMO calculation of entropy has two limitations. It appears that translational entropy was calculated as though the system was gaseous, even though water was specified as a solvent. A matter of future work may be to use the Sackur-Tetrode equation to correct translational entropy so that the calculated units represent the smaller molar volume of a condensed system. In addition,

Table 2 – Aqueous thermodynamic data for macrocycle/ion complexes with MOPAC

Complex	ΔH_{com} (kJ/mol)	ΔS_{com} (J/mol)	ΔG_{com} (kJ/mol)
Nonactin/NaSCN	75.6	-217.2	140.3
Nonactin/KSCN	-152.8	-82.6	-128.2
Tetrafuromacrolide/NaSCN	130.6	-168.3	180.8
Tetrafuromacrolide/KSCN	-72.9	-102.2	-42.5

Table 3 – Aqueous thermodynamic data for macrocycle/ion complexes with NBS values

Complex	ΔH_{com} (kJ/mol)	ΔS_{com} (J/mol)	ΔG_{com} (kJ/mol)
Nonactin/NaSCN	56.7	-126.9	94.9
Nonactin/KSCN	-44.2	-19.2	-38.5
Tetrafuromacrolide/NaSCN	111.8	-77.9	135.0
Tetrafuromacrolide/KSCN	35.7	-38.7	47.2

Table 4 – Difference in Gibbs Free Energy values with KSCN/NaSCN complexes

MSCN from MOPAC/COSMO		MSCN from NBS	
Nonactin	268.5 kJ/mol	Nonactin	133.42 kJ/mol
Tetrafuromacrolide	223.3 kJ/mol	Tetrafuromacrolide	87.8 kJ/mol

COSMO reports that certain internal rotations are ignored during the calculation of entropy which increases the error of the reported entropy value. An additional matter of future work may be to model the solvent-solute interaction in terms of discrete water molecules rather than as a continuum dielectric.

Conclusions

This work examined computational methods to compare thermodynamic values for a proposed complexed ion tetrafuromacrolide with a known ionophore, nonactin. The SPARTAN/MOPAC/COSMO calculations for the free energy of complexation had to be modified to improve the findings for the coordinate covalent bonding between the solvated ions and the organic macrocycle. We used NIST (NBS) values for the solvated ions to replace the calculated values. The NBS modified data showed lower positive changes in Gibb's Free Energy for complexation between the K^+ and Na^+ ligated macrocycles for both nonactin and the proposed tetrafuromacrolide. Nonactin displayed a higher selectivity for the potassium ion than did the proposed compound. Potassium ions are more available for complexation in aqueous solutions due to the high solvation energies of sodium ions. Therefore, the previous results don't necessarily rule out the role of the proposed macrocycle as an ionophore. However, visualization of the optimized complexes does indicate that the nonactin exterior has a more lipophilic nature over the proposed macrocycle. This finding does indicate that the proposed tetrafuromacrocycle would be a less effective ionophore.

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