

Theoretical Study of Stability, Tautomerism, Equilibrium Constants (pK_T) of 2-Thiouracil in Gas phase and Different solvents (water and Acetonitrile) by the Density Functional Theory Method.

N.Surendra Babu

Department of Chemistry, Hawassa University, Hawassa, Post Box No: 5, ETHIOPIA,

Email:nsbabusk@gmail.com, nsbabusk@hu.edu.et, Phone: +251 916160471, Fax.No.+251(046) 2205421

ABSTRACT: A density functional theory (DFT) calculations were carried out to investigate the relative energies, equilibrium constants and dipole moments of 2-thiouracil in the gas phase and in solutions with different polarities. Solvent effects were examined by means of the self-consistent reaction field (SCRF) method. All possible tautomers are optimized at the B3LYP/6-31++G (d,p). All calculations indicate that the most stable tautomer for both species, in the gas phase as well as in solution, has the 2TU1 form. In addition to calculate the thermodynamic properties and the inter conversion equilibrium constants for all tautomers. The molar transfer Gibbs energies of $\Delta\Delta G_t^0$ (solvent1- solvent 2), determines the solvent effect on the position of the equilibrium.

Key words: 2-Thiouracil, DFT method, Tautomerism, relative stabilities, equilibrium constants, and solvent effects.

1. INTRODUCTION

Tautomerism interconversions[1] have been investigated by chemists during last decades. Recently, study of tautomerism received renewed attention due to its importance on determination of compounds' properties and their area of applications. The importance of tautomerism is revealed more since in recent years the investigation about tautomerism has been the major topic in theoretical chemistry. For example, tautomerisms in keto-enol [2], imine-enamine [3] and many other systems [4] have been studied during past decades. Tautomerism and proton transfer of the keto-enol tautomer of heterocyclic systems with several basic centers, N, O and S atoms, are of great interest to medicinal and biochemical applications. Also, understanding of the relative stabilities of heterocyclic tautomers and any subsequent conversions between tautomeric forms is very vital for both structural chemists and biologists [5]. Along this line, relative stabilities of various tautomeric structures of five-, six- and seven-member heterocyclic rings were investigated using both theoretical and experimental tools [6].

In recent years, uracil and its thio derivatives have already become the intense focus of research due to their biological, pharmacological, and spectroscopic activities. For example, a series of thiouracils, such as 2-thiouracil, 4-thiouracil, and 2,4-dithiouracil, have been identified as minor components of t-RNA and peptide nucleic acids, where the former two thiouracils have also been used as anticancer and antithyroid drugs and for the treatment of heart disease [7–11]. The in appropriate replacement of uracil by a thiouracil can be responsible for the misrecognition in m-RNA [12]. Moreover, their various tautomeric forms seem to be important for explaining the mutation occurring during DNA duplication. Meanwhile, the complexes of thiouracil derivatives with transition metal ions of biological importance have been shown to exhibit anticancer and antimicrobial activity [13]. Additionally, thiouracils have also been proposed to be efficient metal corrosion inhibitors [14]. Thus, the properties and characteristics of thiouracils are of great interest in pharmaceutical researches and applications. In the present paper, the geometries, dipole moments, Mulliken charges and relative energies have been calculated, using the DFT method.

2. COMPUTATIONAL METHOD DETAILS

2-thiouracil has 6 keto– enol and enol–enol tautomeric forms altogether. Considering the orientation of the hydroxyl hydrogen and thio hydrogen (it is in the same or opposite direction with the C=N bond of the enol), the number of the tautomers ascends to 12, Fig. [1], illustrates the 12 tautomers. All the geometries of the tautomers have been fully optimized using the B3LYP method within the framework of density functional theory (DFT) in conjunction with the 6-311++G (d,p) basis set, i.e., Becke's three parameter non-local hybrid exchange potential with the non-local correlation functional of Lee, Yang, and Parr [15,16]. The B3LYP method has been shown to yield reliable geometries for a wide variety of systems also including the relevant uracil species [17-19]. Following the geometry optimizations, analytical frequency calculations were proceeded at the DFT/6-311++G(d,p) level, using following standard procedures, to obtain the thermochemical properties. Solvent effect was considered when calculating the equilibrium constant. All these calculations were carried out on a Pentium V personal computer by means of GAUSSIAN09 program package and for our computations [20].

Thermodynamic properties such as: the enthalpy, the Gibbs free energy and the entropy were given according to the formulas [21]

$$H = E + RT \quad (1)$$

$$G = H - TS \quad (2)$$

E is the thermal energy
H is the enthalpy
G is the Gibbs free energy

In order to determine the kinetic parameters of the transformations, we calculated their tautomeric equilibrium constants by using the following relations [22].

$$K_T = \exp(-\Delta G/RT) \quad (3)$$

Where K_T is the equilibrium constant between the tautomers, the gas constant is R is 1.987×10^{-3} kcal.mol⁻¹ and the temperature T is 298.15K. The quantity ΔG is the difference between the Gibbs free energies of the given tautomer with respect to stable one. The pK_T values of the studied molecules were calculated by the following equation [21]:

$$pK_T = -\log K_T \quad (4)$$

Calculated total energies (a.u), thermal Enthalpy (H) (a.u), thermal free energy (G) (a.u), dipole moment (μ) (debye) ,enthalpy change (ΔH^0), free energy change (ΔG^0), entropy change (ΔS^0) equilibrium constants (K) and pK_T for various tautomers. These thermochemical parameters were obtained through the frequency calculation at DFT/6-311++G (d,p) level. The solvents (acetonitrile & water) effect on the tautomerism processes of 2-thiouracil was investigated by self-consistent reaction field (SCRF) method based on Onsager's reaction field theory.

3. RESULTS AND DISCUSSION

3.1. Energies and relative stability

The total and relative energies (at DFT/6-311++G(d,p) level) of 12 tautomers of 2-thiouracil are presented in Table [1], among the 12 tautomers, four are (2TU2, 2TU3, 2TU11,and 2TU12) thio- enols, four are (2TU4, 2TU5, 2TU9,and 2TU10) keto-enols, three are di-enols (keto-enol and thio-enol) and one is keto tautomer (2TU1).The most stable 2TU1 is taken as reference to obtain the relative energetic stability of other tautomers. It can be seen from the results that among four mono-enol tautomers two are thio-enol(2TU11&2TU12) and two are keto-enol (2TU9&2TU10) tautomers have highest energy, with relative energy about 20-25 kcal mol⁻¹ , because of the possibility of migration of hydrogen atoms, and eliminating the simultaneous transfers little likely of more than atom, only the exchange of hydrogen atom between heteroatom spatially close and separate by more than two atoms can be carried out. From the table [1]

shows that the mono-enol tautomers of thio-enol(2TU11&2TU12) and keto-enol (2TU9&2TU10) tautomers have highest energy, with relative energy about 20-25 kcal mol⁻¹. Because of the possibility of migration of hydrogen atoms, and eliminating the simultaneous transfers little likely of more than atom, only the exchange of hydrogen atom between heteroatom spatially close and separate by more than two atoms can be carried out.

Other mono and di-enol tautomers are less relative energy, except the tautomer 2TU5 (20.07kcal/mole), with relative energy about 11–18 kcal mol⁻¹. The energy difference of the tautomers of same tautomeric form that differs in the orientations of hydroxyl hydrogens, varies for specific systems. The order of relative **stabilities** for tautomer in gas phase with respect to stable tautomer: 2TU2 > 2TU6 > 2TU7 > 2TU4 > 2TU3 > 2TU8 > 2TU5 > 2TU12 > 2TU9 > 2TU11 > 2TU10.

Solvent effects are relevant in tautomers stability phenomena, since polarity differences among tautomers can induce significant changes in their relative energies in solution. SCRF/B3LYP calculations were used to analyze the solvent effects on tautomerism of 2-thiouracil. The data presented in Table [1] show that polar solvents increase the stability of all 2-thiouracil tautomers in compare to gas phase. The difference between the total energies of 2-thiouracil and the other forms do not show a regular trend when changing from gas phase to most polar solvents (water). The solvent interactions have pronounced effect on the order of stability of the tautomers in the gas phase. The order of stability of 2-thiouracil in acetonitrile: 2TU5> 2TU2 > 2TU3 > 2TU12 > 2TU6 > 2TU7 > 2TU11 > 2TU9 > 2TU8 > 2TU10 > 2TU4 and The order of stability in water: 2TU4> 2TU5 > 2TU2 > 2TU3 > 2TU12 > 2TU6> 2TU11 > 2TU7 > 2TU9 > 2TU8 > 2TU10.

Th dipole moment (μ) has been calculated in different media, at the same basis set level and presented in Table [2]. It seems that the dipole moments (μ) increases by changing the gas phase to the solution as well as by increasing the solvent polarity. On going from gas phase to solution, dipole **moments** of all isomers are increased, as the solvent becomes much more polar. The increase in dipole **moments** implies that charges around the molecule are probably more developed due to the polarization induced by solvents.

3.2 The thermodynamic parameters

The thermodynamic parameters are presented in Table [3] in different media, the ΔH , ΔG and ΔS values are all positive for different solvents except 2TU1 $\leftrightarrow$ 2TU4 has negative ΔS value in all solvents. ΔH values are all positive, i.e. endothermic reaction, ΔS usually is relatively small, indicate that with no ring formation or other unusual changes in the constraints between the products and reactants.

The effect of the medium on the position of equilibrium can be considered from two points of view: (a) comparison of the gas-phase and solution equilibrium constants, and (b) comparison of the equilibrium constants for different solvents. These are primarily non-ionic reactions where the interaction between reacting molecules and solvent is relatively small. Let us consider a simple isomerization reaction $A \leftrightarrow B$ in the solvents I and II, whose abilities to solvate A and B are different. This corresponds to the Gibbs energy diagram shown in Fig. [2] [23]. From Fig. [2], Eq. (5) can be immediately derived,

$$\Delta G^0(\text{II}) + \Delta G_t^0(A, \text{I} \rightarrow \text{II}) = \Delta G_t^0(B, \text{I} \rightarrow \text{II}) = \Delta G_t^0(B, \text{I} \rightarrow \text{II}) + \Delta G^0(\text{I}) \quad (5)$$

which, on rearrangement, leads to Eq.6

$$\begin{aligned} \Delta G^0(\text{II}) - \Delta G^0(\text{I}) &= \Delta G_t^0(B, \text{I} \rightarrow \text{II}) - \Delta G_t^0(B, \text{I} \rightarrow \text{II}) \\ &= \Delta \Delta G^0(\text{I} \rightarrow \text{II}) \end{aligned} \quad (6)$$

Since, for equilibria, the logarithm of the equilibrium constant is proportional to the standard molar Gibbs energy change, ΔG^0 , according to Eq. (7),

$$\Delta G^0 = -RT \ln K \quad (7)$$

It follows from Eqs. (6) and (7) that the difference in the molar transfer Gibbs energies of reactant A and product B, $\Delta \Delta G_t^0(\text{I} \rightarrow \text{II})$, determines the solvent effect on the position of this equilibrium. In studying solvent effects on equilibria, it is, in principle, not sufficient to investigate the ΔG^0 changes alone, because this term is determined by both an enthalpy and an entropy term according to Eq. (8).

$$\Delta G^0 = \Delta H^0 - T\Delta S^0 \quad (8)$$

Transfer functions can also be defined for the thermodynamic state functions ΔH^0 and ΔS^0 . If both transfer Gibbs energies and transfer enthalpies are available, it should be possible to achieve a complete dissection of the effect of solvents on the various thermodynamic parameters. The values of $\Delta \Delta H^0$ and $T \Delta \Delta H^0$ were presented in Table [4] in all the solvents. From the table four types of reaction control can be recognized:

(a) Cooperative effects, with $\Delta\Delta H^0$ and $T\Delta\Delta S^0$ having opposite signs. Then these two terms will be additive; (b) Enthalpy-controlled effects, in which the two terms are opposed, but the enthalpy term is larger; (c) Entropy-controlled effects, in which the two terms are opposite but the $\Delta\Delta S^0$ is larger; and (d) Compensating effects, in which the two terms are opposed but nearly equal.

3.3. Equilibrium constants

In order to determine the kinetic parameters of the transformations, we calculated their tautomeric equilibrium constants by using the relation [21]. The constants were calculated by considering that all the tautomers can be in the equilibrium with the most stable tautomer. Table [5] contained the equilibrium constants (K) and the pK_T values in different solvents, calculated from tautomeric most stable which was 2TU1. All pK_T values in gas phase and the two solvents were positive and this confirmed the fact that 2TU1 tautomer is most stable.

Calculated values for the equilibrium between the tautomers of 2-thiouracil show that the 2TU1 is more dominant than the 2TU2, for both the gas and solutions (water and acetonitrile) with pK_T value of 6.5874, 8.4344 and 8.3833, respectively. The 2TU1 form is also a more dominant tautomer than the 2TU6 form, 2TU7 form, 2TU3 form and the 2TU4 form. Tautomers 5, 8, 9, 10, 11 and 12 are not present in detectable amounts. In the gas phase the order of the stability of 2-thiouracil tautomers is $1 > 6 > 7 > 3 > 4$ while tautomers 5, 8, 9, 10, 11 and 12 are not detected. However, we predict the order of stability in aqueous and acetonitrile solutions are same, to be $1 > 4 > 2 > 3 > 5$.

We can return to our diagram of interconversion, to evaluate in a more realistic way the constants of equilibrium as presented in Fig [3]. Table [6] contained the pK_T of the equilibrium constants calculated from the general outline of interconversion. Some pK_T were positive and other negative; that determine the privileged direction of equilibrium. If the pK_T was positive, equilibrium moved from right towards the left and when it was negative, equilibrium moved from left towards the right.

4. Conclusion

For 2-thiouracil, tautomerization in gas phase and solution was studied using DFT at the B3LYP/6-31++G(d,p) basis set. In this work we calculated relative stability, the constants of equilibrium for different tautomers of 2-thiouracil. According to our calculations, the most stable form is di ketone form (2TU1) and

the order of energies; acetonitrile > water > gas, the dipole movements are order: Gas > acetonitrile > water, because of polarity increasing. In addition the equilibrium constants and inter conversation equilibrium constants also calculated. Form this we observed some pK_T were positive and other negative; that determine the privileged direction of equilibrium.

REFERENCES

1. Belova N V, Oberhammer H, Girichev G V, Shlykov S A. Tautomeric Properties and Gas-Phase Structure of 3-Chloro-2,4-pentanedione. J Phys Chem A. 2008;112(14): 3209-3214.
2. Misra A, Dalai S. A DFT-based comparative study on the excited states intramolecular proton transfer in 1-hydroxy-2-naphthaldehyde and 2-hydroxy-3-naphthaldehyde.J Mol Struct. (Theochem.). 2007;807: 33-41.
3. Dines T, Onoh H. An infrared and resonance Raman spectroscopic study of phenylazonaphthol pigments. Spectrochim Acta A. 2006; 64A: 891-900.
4. Bonacin J A, Formiga A L B, de Melo V H S, Toma H E. Vibrational spectra and theoretical studies of tautomerism and hydrogen bonding in the violuric acid and 6-amino-5-nitrosouracil system.Vib Spectrosc. 2007;44(1):133-141
5. Elguero J, Katritzky A R, Denisko O V. Prototropic tautomerism of heterocycles: Heteroaromatic tautomerism—General overview and methodology.Adv Heterocycl Chem. 2000;76: 1-85.
6. Santos M, Junior M, Oliveira S, da Silva J, Lima M, Lima M, Galdino S, Pitta I. Configurational and conformational analysis of some fully substituted imidazolidinic compounds: a theoretical study.J Mol Struct., (Theochem). 2005;715: 191-198.
7. Sulkowska A, Rownicka J, Bojko B, Sulkowski W. Interaction of anticancer drugs with human and bovine serum albumin.J Mol Struct.2003; 651:133-140.
8. Theodossiou C, Schwarzenberger PA . Theodossiou C, Schwarzenberger P. Propylthiouracil reduces xenograft tumor growth in anathymic nude mouse prostate cancer model.Am J Med Sci. 2000; 319:96
9. Elias AN. Anti-thyroid thioureylenes in the treatment of psoriasis. Med Hypotheses. Med Hypotheses. 2004; 62:431-437.
10. Antoniadis CD, Corban GJ, Hadjikakou SK, Hadjiliadis N, Kubicki M, Warner S, Butler IS . Synthesis and Characterization of (PTU)₂ (PTU = 6-n-propyl-2-thiouracil) and (CMBZT)₂ (CMBZT = 5-chloro-2-mercaptobenzothiazole) and Possible Implications for the Mechanism of Action of Anti-Thyroid Drugs. Eur J Inorg Chem. 2003; 8:1635-1640.
11. Imam DR, El-Barbary AA, Nielsen C, Pedersen EB. Anti-HIV active naphthyl analogues of HEPT and DABO.Monatsh Chem.2002; 133:723-732.
12. Beck CF, Howlett G.J Mol Biol. The nature of the miscoding caused by growth in the presence of 2-thiouracil.1977; 111:1-17.
13. Kamalakannan P, Venkappayya D. . Synthesis and characterization of cobalt and nickel chelates of 5-dimethylaminomethyl-2-thiouracil and their evaluation as antimicrobial and anticancer agents J Inorg Biochem.2002; 90:22-37.
14. Dafali A, Hammouti B, Mokhlisse R, Kertit S. Corros Sci. Substituted uracils as corrosion inhibitors for copper in 3% NaCl solution.2003; 45:1619-1628.
15. Becke AD. Density-functional thermochemistry. III. The role of exact exchange J Chem Phys.1993;98:5648-5653.
16. Lee C, Yang W, Parr RG.Development of the Colle-Salvetti correlation-energy formula into a functional of the electron density. Phys Rev B.1988;37:785-793.
17. Lamsabhi AM, Alcami M, Mo O, Yanez M, Tortajada. Association of Cu²⁺ with Uracil and Its Thio Derivatives: A Theoretical Study J.ChemPhysChem. 2004;5:1871-1878.

18. Lamsabhi AM, Alcamí M, Mo O, Yanez M. Gas-Phase Reactivity of Uracil, 2-Thiouracil, 4-Thiouracil, and 2,4-Dithiouracil towards the Cu⁺ Cation: A DFT Study *ChemPhysChem*. 2003; 4:1011-1016.
19. Llamas-Saiz AL, Foces-Foces C, Mo O, Yanze M, Elguero E, Elguero J. The geometry of pyrazole: A test for ab initio calculations. *J Comput Chem*. 1995; 16:263-272.
20. Frisch J, Trucks GW, Schlegel HB, Scuseria GE, Robb MA, Cheeseman JR, Scalmani G, Barone V, Mennucci B, Petersson GA, Nakatsuji H, Caricato M, Li X, Hratchian HP, Izmaylov AF, Bloino J, Zheng G, Sonnenberg JL, Hada M, Ehara M, Toyota K, Fukuda R, Hasegawa J, Ishida M, Nakajima T, Honda Y, Kitao O, Nakai H, Vreven T, Montgomery JA, Peralta Jr. JE, Ogliaro F, Bearpark M, Heyd JJ, Brothers E, Kudin KN, Staroverov VN, Keith T, Kobayashi R, Normand J, Raghavachari K, Rendell A, Burant J. C, Iyengar SS, Tomasi J, Cossi M, Rega N, Milla JM, Klene, M, Knox JE, Cross JB, Bakken V, Adamo C, Jaramillo J, Gomperts R, Stratmann R. E, Yazyev O, Austin AJ, Cammi R, Pomelli C, Ochterski JW, Martin RL, Morokuma K, Zakrzewski VG, Voth GA, Salvador P, Dannenberg JJ, Dapprich S, Daniels AD, Farkas O, Foresman JB, Ortiz JV, Cioslowski J, and Fox DJ, Gaussian, Inc. Wallingford CT. Gaussian 09. 2010; Revision B.01,M.
21. Assoma A, Benjamine, Bede A. Lucie, Kone M, N'Gussan Y. Thomas. Theoretical study of stability, tautomerism, equilibrium constants (pK_T), Activation energies and acidity of 6-thioxanthine in gas phase by the AB initio method and functional density theory calculations. *European journal of scientific research*. 2010; 44(2): 337-354.
22. Peng C, Ayala P. Y, Schlegel HB, Frisch M. J. Using redundant internal coordinates to optimize equilibrium geometries and transition states. *J. Comp. Chem*. 1996; 17: 49-56.
23. *Solvents and Solvent Effects in Organic Chemistry, Fourth Edition*. Edited by Christian Reichardt and Thomas Welton Copyright 2011 WILEY-VCH Verlag GmbH & Co. KGaA, Weinheim.