



Importance of CH tautomers in the tautomeric mixture of uric acid

E.D. Raczyńska^{a,*}, M. Makowski^b, M. Szeląg^c, B. Kamińska^a, K. Zientara^c

^a Department of Chemistry, Warsaw University of Life Sciences (SGGW), ul. Nowoursynowska 159c, 02-776 Warszawa, Poland

^b Faculty of Chemistry, University of Gdańsk, ul. Sobieskiego 18, 08-952 Gdańsk, Poland

^c Interdisciplinary Department of Biotechnology, Warsaw University of Life Sciences (SGGW), ul. Nowoursynowska 166, 02-776 Warszawa, Poland

ARTICLE INFO

Article history:

Received 17 December 2009

Received in revised form 29 January 2010

Accepted 30 January 2010

Available online 10 February 2010

Keywords:

Uric acid

UA building blocks (purine, uracil, 2-imidazolone)

Stability of CH tautomers

Intramolecular interactions

π -Electron delocalization

Tautomeric equilibria

ABSTRACT

To understand the formation and degradation processes of uric acid (**UA**) in living organisms we considered all possible tautomers–rotamers of **UA** by semiempirical (AM1) method, and next we used the *ab initio* (HF, MP2, and G2) and DFT methods to the most stable five isomers of **UA** selected at the AM1 level. Our calculations show evidently that at least two CH–NH forms should be considered in the tautomeric mixture of **UA** to understand the formation of the intermediate 5-hydroxyisourate (HIU) upon the **UA** oxidation by uricase. Intramolecular interactions and π -electron delocalization for **UA** are very similar to those occurring for its building blocks (purine (**P**), uracil (**U**), and 2-imidazolone (**I**)). Tautomeric equilibrium constants for NH \rightarrow OH, NH \rightarrow CH, and OH \rightarrow CH conversions in **UA** are close to those for **U** and **I**. Tautomeric equilibrium constants for NH \rightarrow NH conversions in **UA** are close to those for **P**. Favored tautomers for **UA**, **U** and **I** are strongly stabilized by intramolecular interactions between the amide groups, whereas that for **P** is stabilized by π -electron delocalization and $N^9H \cdots N^3$ intramolecular interaction. Aromaticity seems to be very important factor that influences the tautomeric preference for the systems which do not possess the *exo* functional groups.

© 2010 Elsevier B.V. All rights reserved.

1. Introduction

Uric acid (**UA**) – isolated from human urine more than 200 years ago [1] – presents very complex physicochemical, chemical, bio-chemical and biological properties, which have been extensively studied during the last five decades. As one of the end products of the metabolism for purine nucleotides in humans [2], **UA** causes various opposite effects on human health, positive effects for substantial concentrations (antioxidation, anticarcerogen properties, and longevity) and negative effects for elevated quantities in blood, tissues and urine (hyperuricemia) [3].

Similar to building blocks (Fig. 1): purine (**P**), uracil (**U**), and 2-imidazolone (**I**), **UA** exhibits prototropic tautomerism [4,5]. It possesses four hydrogens that can move as protons between seven heteroatoms (three *exo*-oxygen and four *endo*-nitrogens) and also between heteroatoms and five *endo*-carbon atoms. Consequently, combinations of various types of tautomeric conversions in **UA** such as amide-iminol, keto-enol, amine-imine, and/or imine-enamine lead to 24 NH–OH tautomers and 50 CH–NH–OH tautomers (Table 1). Solely in the solid state, the structures of dihydrate and anhydrous **UA** and its salts (urates) have been experimentally determined [6,7]. Urates in the solid state are mainly monoanions [7]. Uric acid in the solid state takes the tetra-NH (tri-oxo) form

(**UA1**, Table 1) with protons at the N^1 , N^3 , N^7 , and N^9 atoms [6]. The **UA1** form seems to be also favored in the gas phase and in solution [8,9].

Experimental investigations of prototropy for heterocycles are exceptionally difficult to carried out, because tautomeric conversions are very fast processes, and also, because most of experimental techniques are incapable to detect less than 0.1% of minor tautomers [4]. Fortunately, quantum-chemical methods can be applied even for very complicated systems. These methods give the possibilities to optimize the structure of all possible tautomeric forms and to study all tautomeric equilibria. They make also possible to estimate thermodynamic parameters such as the enthalpy (H_T), entropy (S_T), Gibbs free energy (G_T), tautomeric equilibrium constant (pK_T) for each tautomeric conversion, and the percentage content of each tautomer in the tautomeric mixture.

To our knowledge [10], all 74 tautomers possible for **UA** have been never considered in the literature [8,9]. Shukla and Mishra [8] chose only three NH–OH forms for their semiempirical calculations (MNDO, AM1, and CNDO/s-CI). Demir et al. [9a] reported the semiempirical (MNDO, AM1, and PM3) results for 15 NH–OH tautomers. The same isomers were considered by Monard and co-workers [9b,c] who used the semiempirical (AM1 and PM3), *ab initio* (HF and MP2), and DFT methods. Using the HF and DFT methods, Jiménez and Alderete [9d] analysed 16 NH–OH forms. Leszczynski and co-workers [9e] reported the DFT results for 18 NH–OH isomers. Twenty-four NH–OH tautomers and additionally

* Corresponding author. Tel.: +48 22 5937623.

E-mail address: ewa_raczynska@sggw.pl (E.D. Raczyńska).

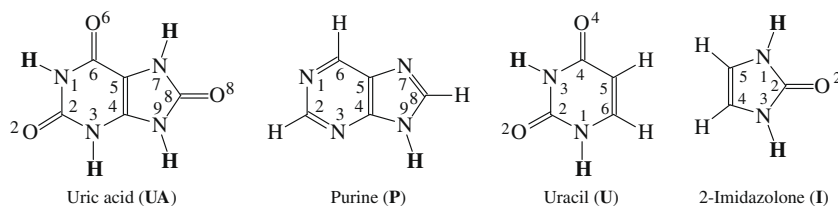


Fig. 1. Uric acid (UA) and its building blocks: purine (P), uracil (U), and 2-imidazolone (I). Moving protons are marked in bold.

Table 1
Positions of moving protons for prototropic tautomers of uric acid.

Isomer ^a	Type of tautomer	Position of H	Isomer ^a	Type of tautomer	Position of H
UA1	Tetra-NH	N1N3N7N9	UA38a,b	Mono-CH-di-NH-mono-OH	C6N1N3O6
UA2a,b	Tri-NH-mono-OH	N1N7N9O2	UA39a,b	Mono-CH-di-NH-mono-OH	C6N1N9O6
UA3a,b	Tri-NH-mono-OH	N3N7N9O2	UA40a,b	Mono-CH-di-NH-mono-OH	C5N1N3O8
UA4a,b	Tri-NH-mono-OH	N1N3N7O6	UA41a,b	Mono-CH-di-NH-mono-OH	C4N1N3O8
UA5a,b	Tri-NH-mono-OH	N1N7N9O6	UA42a,b	Mono-CH-di-NH-mono-OH	C5N1N7O8
UA6a,b	Tri-NH-mono-OH	N3N7N9O6	UA43a,b	Mono-CH-di-NH-mono-OH	C5N1N9O8
UA7a,b	Tri-NH-mono-OH	N1N3N7O8	UA44a,b	Mono-CH-di-NH-mono-OH	C8N1N3O8
UA8a,b	Tri-NH-mono-OH	N1N3N9O8	UA45a,b	Mono-CH-di-NH-mono-OH	C8N1N9O8
UA9a-d	Di-NH-di-OH	N1N7O2O6	UA46a-d	Mono-CH-mono-NH-di-OH	C5N7O2O6
UA10a-d	Di-NH-di-OH	N3N7O2O6	UA47a-d	Mono-CH-mono-NH-di-OH	C4N9O2O6
UA11a-d	Di-NH-di-OH	N7N9O2O6	UA48a-d	Mono-CH-mono-NH-di-OH	C2N3O2O6
UA12a-d	Di-NH-di-OH	N1N7O2O8	UA49a-d	Mono-CH-mono-NH-di-OH	C2N9O2O6
UA13a-d	Di-NH-di-OH	N1N9O2O8	UA50a-d	Mono-CH-mono-NH-di-OH	C6N1O2O6
UA14a-d	Di-NH-di-OH	N3N7O2O8	UA51a-d	Mono-CH-mono-NH-di-OH	C6N3O2O6
UA15a-d	Di-NH-di-OH	N3N9O2O8	UA52a-d	Mono-CH-mono-NH-di-OH	C6N9O2O6
UA16a-d	Di-NH-di-OH	N1N3O6O8	UA53a-d	Mono-CH-mono-NH-di-OH	C5N1O2O8
UA17a-d	Di-NH-di-OH	N1N7O6O8	UA54a-d	Mono-CH-mono-NH-di-OH	C4N1O2O8
UA18a-d	Di-NH-di-OH	N1N9O6O8	UA55a-d	Mono-CH-mono-NH-di-OH	C5N3O2O8
UA19a-d	Di-NH-di-OH	N3N7O6O8	UA56a-d	Mono-CH-mono-NH-di-OH	C4N3O2O8
UA20a-d	Di-NH-di-OH	N3N9O6O8	UA57a-d	Mono-CH-mono-NH-di-OH	C5N7O2O8
UA21a-h	Mono-NH-tri-OH	N1O2O6O8	UA58a-d	Mono-CH-mono-NH-di-OH	C5N9O2O8
UA22a-h	Mono-NH-tri-OH	N3O2O6O8	UA59a-d	Mono-CH-mono-NH-di-OH	C2N1O2O8
UA23a-h	Mono-NH-tri-OH	N7O2O6O8	UA60a-d	Mono-CH-mono-NH-di-OH	C8N1O2O8
UA24a-h	Mono-NH-tri-OH	N9O2O6O8	UA61a-d	Mono-CH-mono-NH-di-OH	C8N3O2O8
UA25	Mono-CH-tri-NH	C5N1N3N7	UA62a-d	Mono-CH-mono-NH-di-OH	C8N9O2O8
UA26	Mono-CH-tri-NH	C5N1N7N9	UA63a-d	Mono-CH-mono-NH-di-OH	C5N3O6O8
UA27	Mono-CH-tri-NH	C4N1N3N9	UA64a-d	Mono-CH-mono-NH-di-OH	C4N3O6O8
UA28a,b	Mono-CH-di-NH-mono-OH	C5N1N7O2	UA65a-d	Mono-CH-mono-NH-di-OH	C5N7O6O8
UA29a,b	Mono-CH-di-NH-mono-OH	C4N1N9O2	UA66a-d	Mono-CH-mono-NH-di-OH	C5N9O6O8
UA30a,b	Mono-CH-di-NH-mono-OH	C5N3N7O2	UA67a-d	Mono-CH-mono-NH-di-OH	C6N1O6O8
UA31a,b	Mono-CH-di-NH-mono-OH	C4N3N9O2	UA68a-d	Mono-CH-mono-NH-di-OH	C8N3O6O8
UA32a,b	Mono-CH-di-NH-mono-OH	C5N7N9O2	UA69a-d	Mono-CH-mono-NH-di-OH	C8N9O6O8
UA33a,b	Mono-CH-di-NH-mono-OH	C2N1N3O2	UA70a-h	Mono-CH-tri-OH	C5O2O6O8
UA34a,b	Mono-CH-di-NH-mono-OH	C2N1N9O2	UA71a-h	Mono-CH-tri-OH	C4O2O6O8
UA35a,b	Mono-CH-di-NH-mono-OH	C5N3N7O6	UA72a-h	Mono-CH-tri-OH	C2O2O6O8
UA36a,b	Mono-CH-di-NH-mono-OH	C4N3N9O6	UA73a-h	Mono-CH-tri-OH	C6O2O6O8
UA37a,b	Mono-CH-di-NH-mono-OH	C5N7N9O6	UA74a-h	Mono-CH-tri-OH	C8O2O6O8

^a Rotamers of the OH group(s) are named as **a, b**, etc.

11 zwitterionic structures were considered by Chen et al. [9f] who applied the AM1, HF, and DFT methods. The zwitterionic structures, however, cannot be treated as prototropic tautomers because of charge separation.

To well understand the physicochemical, chemical, biochemical, and biological properties of uric acid, particularly its formation and degradation processes in living organisms, we considered here all possible 74 tautomers of UA (Table 1) and their different conformations (Fig. S1, Supplementary material). First, we used the semiempirical Austin Model 1 (AM1), introduced by Dewar et al. [11] to study the proton-transfer reactions [12]. Next, to well establish the stability order of the most stable five forms of UA selected at the AM1 level, we used the *ab initio* (HF, MP2, and G2) and DFT methods [13]. The AM1 method reproduces well the experimental data for model compound of nucleobases, 2-hydroxypyridine (OH form) → 2-pyridone (NH form) [14,15]. It also predicts the amounts of the OH and NH forms similar to those calculated at the Gaussian-2 (G2) level, recommended for the proton-transfer

reactions in the gas phase [16]. We analysed tautomeric conversions, intramolecular interactions and π -electron delocalization for UA, and we compared them with those occurring for its building blocks (P, U, and I).

2. Methods

2.1. Quantum-chemical calculations

All possible tautomers–conformers for uric acid (Fig. S1, Supplementary material) and all possible tautomers–conformers for building blocks (Schemes S1–S3, Supplementary material), purine, uracil, and 2-imidazolone, were considered and the heats of formations (H_f at 298.15 K) calculated using the semiempirical (AM1) method [11] and the HyperChem program [17]. High level quantum-chemical calculations were also performed for five isomers of UA, selected at the AM1 level, using the HF, MP2, G2, and

Download English Version:

<https://daneshyari.com/en/article/7486488>

Download Persian Version:

<https://daneshyari.com/article/7486488>

[Daneshyari.com](https://daneshyari.com)