Amino Acids (2009) 36:195–201
DOI 10.1007/s00726-008-0047-4

ORIGINAL ARTICLE

Synthesis, spectroscopic and structural elucidation of tyrosinamide hydrogensquarate monohydrate

**Tsonko M. Kolev · Bojidarka B. Koleva ·
Michael Spiteller · William S. Sheldrick ·
Heike Mayer-Figge**

Received: 1 December 2007 / Accepted: 11 February 2008 / Published online: 4 March 2008

Abstract Synthesis, isolation, spectroscopic and structural elucidation of tyrosinamide hydrogensquarate monohydrate (**I**) is reported on the basis of quantum chemical DFT calculations, vibrational analysis and experimental linear-polarized IR-spectroscopy in solid state. These data are compared with those obtained using single crystal X-ray diffraction, which show that the molecules of (**I**) in the unit cell formed 3D network through moderate intermolecular $_{(Tyr)}OH\cdots O=C_{(Sq)}$ (2.727 Å), $O{=}C{-}NH_2\cdots OH_{(Tyr)}$ (2.991 Å), $O{=}C{-}NH_2\cdots OH_{(Sq)}$ (2.988 Å), $O{=}C{-}NH_2\cdots O{=}C{-}NH_2$ (3.068 Å), $N^+H_3\cdots O{=}C_{(Sq)}$ (2.737, 2.953, 2.954 Å), $OH_2\cdots O{=}C_{(Sq)}$ (2.839 Å) and $_{(Sq)}OH\cdots OH_2$ (2.607 Å) hydrogen bonds. The relationship between the structure and spectroscopic properties is studied.

Keywords Tyrosinamide hydrogensquarate monohydrate · IR-LD spectroscopy · DFT calculations · Vibrational analysis · Single crystal X-ray diffraction · FAB-MS

Introduction

The important biorole of squaric acid, its amides and other derivatives was intensively studied in the previous years. Some of them are effective monoanionic inhibitors of protein tyrosine phosphatases[1], selective inhibitors of DNA polymerases (Xie et al. 2004), potential antitumor agents (Tevyashova et al. 2004; Sztaricskai et al. 2005), selective antagonist of ionotropic glutamate receptors (Xie et al. 2004; Tevyashova et al. 2004; Sztaricskai et al. 2005), potential potassium channel openers (Gilbert et al. 2000), medications for treatment of asthma, multiple sclerosis and rheumatoid arthritis (Porter et al. 2000) or inhibitors of matrix metalloproteases (Onaran et al. 2005). The biological activity of tyrosinamide could be illustrated with its participation in the novel thermoresponsive polymeric drug delivery system with ended L-tyrosinamide group, designed for local radiotherapy (Hruby et al. 2005) or with its structural role in hAGRP-melanocortin agonist peptide template Tyr-c[*β*-Asp-His-DPhe-Arg-Trp-Asn-Ala-Phe-Dpr]-Tyr-NH_2 (Wilczynski et al 2005), a power like decay function examining the fluorescence decay of tyrosine (L-tyrosine and *N*-acetyl-L-tyrosinamide) (Marx et al. 2004) or with its structure-based drug design as a new serotonin derivative, serotonin–*O*–carboxymethyl-glycyl-tyrosinamide, for the various $(5\text{-HT})_1$ receptor subtypes (Vianini et al 2001). Therefore, synthesis, isolation, spectroscopic and structural characterization of tyrosinamidium hydrogensquarate monohydrate (**I**) is reported (Scheme 1).

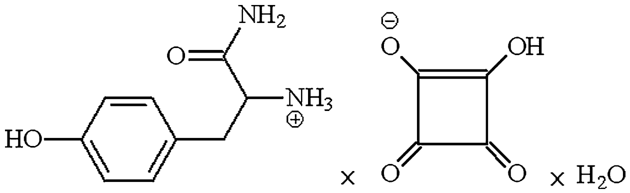
T. M. Kolev (✉)
Bulgarian Academy of Sciences, Institute of Organic Chemistry,
Acad. G. Bonchev Str. build. 9, 1113 Sofia, Bulgaria
e-mail: kolev@orgchm.bas.bg

T. M. Kolev
Faculty of Chemistry, Department of Organic Chemistry,
Plovdiv University "P. Hilendarski", 24 Tzar Assen Str,
4000 Plovdiv, Bulgaria

B. B. Koleva
Faculty of Chemistry, Department of Analytical Chemistry,
Sofia University "St. Kl. Ohridsky, 1164 Sofia, Bulgaria

M. Spiteller
Institut für Umweltforschung, Universityat Dortmund,
Otto-Hahn-Strasse 6, 44221 Dortmund, Germany

W. S. Sheldrick · H. Mayer-Figge
Lehrstuhl für Analytische Chemie, Ruhr-Universitat Bochum,
University Strasse 150, 44780 Bochum, Germany



Scheme 1 Chemical diagram of tyrosinamidium hydrogensquarate monohyfrate

Basing on the fact that the amino acid amides are more than 40,000 biologically active (Merkler 1994; Suwan et al. 1994; Kulathila et al. 1999) compounds, comparing with corresponding free acids, the expectance of its compound a pharmaceutical application is reasonable.

Experimental Part

Synthesis

Tyrosinamide hydrogensquarate monohydrate was synthesized by adding water solution of tyrosinamide (0.4450 g) to 1 M solution of squaric acid (0.4321 g, H_2O) with continuous stirring and heating. A colorless precipitate was obtained from the resulting solution after 30 min. The product was filtered after cooling and recristallized from methanol–water (1:1) to afford the compound in quantitative yield. The purity of the compound was confirmed by IR and UV-Vis spectroscopy. Single prismatic and colorless crystals, suitable for X-ray analyses, were grown from methanol–water (1:1) at room temperature over a period of 2 weeks. (Found: C, 50.01; H, 5.16; N, 8.98; $[C_{13}H_{16}N_2O_7]$ calcd.: C, 50.00; H, 5.16; N, 8.97 %). The most intensive signal in the mass spectrum is that of the peak at 181.25 *m/z*, corresponding to the singly charged cation $[C_9H_{13}N_2O_2]^+$ with a molecular weight of 181.21. The last data indicated synonymously the synthesis and isolation of titled compound.

Materials and methods

Tyrosinamide and squaric acid were purchased from Sigma-Aldrich and Merck, respectively.

The X-ray diffraction intensities were measured in the ω scan mode on a Siemens P4 diffractometer equipped with Mo K_α radiation ($\lambda = 0.71073$ Å $\theta_{\max} = 30°$). The structure was solved by direct methods with displacement ellipsoids depicted (Sheldrick 1995) and refined against F^2 (Sheldrick 1997). An ORTEP plot illustrates the anion and cation structures at 50% probability level. Relevant crystallographic structure data and refinement details are presented in Table 1, and selected bond distances and angles in Table 2. The hydrogen atoms were constrained to calculated positions and refined using riding models in all cases.

Table 1 Crystal data, data collection and refinement results for tyrosinamidium hydrogensquarate monohydrate

Empirical formula	C_{13} H_{16} N_2 O_7	
Formula weight	312.28	
Temperature (K)	293(2)	
Wavelength (Å)	0.71073	
Crystal system, space group	Monoclinic, *P2₁*	
Unit cell dimensions	$a = 4.883(4)$ Å	$\beta = 98.74(6)°$
	$b = 11.353(2)$ Å	
	$c = 12.284(4)$ Å	
Volume (Å^3)	673.1(7)	
Z	2	
Calculated density (Mg m^{-3})	1.541	
Absorption coefficient (mm^{-1})	0.127	
F(000)	328	
Crystal size (mm)	0.59, 0.34, 0.34	
θ Range for data collection	$7.5 \leq \theta \leq 15.0$	
Limiting indices	$-6 \leq h \leq 1$, $-15 \leq k \leq 1$, $-17 \leq l \leq 17$	
Absorption correction	ψ Scans	
Goodness-of-fit on F^2	1.085	
Final *R* indices [$I > 2\sigma$ (I)]	$R1 = 0.0486$, $wR2 = 0.1105$	
R indices (all data)	$R1 = 0.0695$, $wR2 = .1240$	

The IR-spectra were measured on a Bomem–Michelson 100 FTIR-spectrometer (4,000–400 cm^{-1}, 2 cm^{-1} resolution, 150 scans) equipped with a Perkin Elmer wire-grid polarizer. Non-polarized solid-state IR spectra were recorded using KBr disk technique. The oriented samples were obtained as a suspension in a nematic liquid crystal (MLC 6815, Merck) with the presence of an isolated nitrile stretching IR-band at about 2,230 cm^{-1}, additionally serving as an orientation indicator. The validation of this new orientation solid-state method [published first in 2004 (Ivanova et al. 2004)], used in linear-dichroic infrared (IR-LD) spectroscopy, based on suspension in nematic liquid crystal for accuracy and precision, influence of the liquid crystal medium on peak positions and integral absorbances of the guest molecule bands have been presented (Ivanova et al. 2006a, b, 2007). Optimization of experimental conditions and an experimental design for quantitative evaluation of the impact of four input factors have been presented (Ivanova et al. 2006a, b, 2007). The number of scans, the rubbing-out of KBr-pellets, the amount of studied compounds included in the liquid crystal medium and the ratios of Lorentzian to Gaussian peak functions in the curve fitting procedure on the spectroscopic signal at five different frequencies have been

Table 2 Selected bond lengths (Å) and angles (°) for tyrosynamide hydrogensquarate monohydrate from crystallographic data

C1 O1 1.233(4)	O1 C1 N1 124.7(3)
C1 N1 1.313(4)	O1 C1 C2 119.2(3)
C1 C2 1.526(4)	N1 C1 C2 116.0(3)
C2 N2 1.483(4)	N2 C2 C1 107.6(3)
C2 C3 1.545(4)	N2 C2 C3 111.5(3)
C3 C11 1.495(4)	C1 C2 C3 112.1(3)
C11 C16 1.391(5)	C16 C11 C12 117.8(3)
C11 C12 1.393(5)	C16 C11 C3 120.5(3)
C12 C13 1.380(5)	C12 C11 C3 121.7(3)
C14 O14 1.356(4)	C13 C12 C11 121.5(3)
C14 C15 1.394(5)	C12 C13 C14 119.5(3)
C15 C16 1.381(5)	O14 C14 C13 118.1(3)
C1′ O1′ 1.229(4)	O14 C14 C15 121.6(3)
C1′ C2′ 1.472(5)	C13 C14 C15 120.3(3)
C1′ C4′ 1.495(5)	C16 C15 C14 119.0(3)
C2′ O2′ 1.250(4)	O1′ C1′ C2′ 133.7(3)
C2′ C3′ 1.433(5)	O1′ C1′ C4′ 137.8(3)
C3′ O3′ 1.310(4)	C2′ C1′ C4′ 88.5(3)
C3′ C4′ 1.430(4)	O2′ C2′ C3′ 137.4(3)
C4′ O4′ 1.239(4)	O2′ C2′ C1′ 132.8(3)
OW HW1 0.830(11)	C3′ C2′ C1′ 89.8(3)
OW HW2 0.828(10)	O3′ C3′ C4′ 135.1(3)
	O3′ C3′ C2′ 132.2(3)
	C4′ C3′ C2′ 92.7(3)
	O4′ C4′ C3′ 136.0(3)
	O4′ C4′ C1′ 135.0(3)
	C3′ C4′ C1′ 89.0(3)
	HW1 OW HW2 126(5)

studied (Ivanova et al. 2006a, b, 2007). It has been found that the procedure for the position (v_i) and integral absorbancies (A_i) determination for each i-peak has been carried out by deconvolution and curve-fitting procedures at 50:50% ratio of Lorentzian to Gaussian peak functions, χ^2 factors within 0.00066–0.00019 and 2,000 iterations. The means of two treatments were compared by Student *t* test. The experimental IR-spectral patterns have been acquired and processed by GRAMS/AI 7.01 IR spectroscopy (Thermo Galactic, USA) and STATISTICA for Windows 5.0 (StatSoft, Inc., Tulsa, OK, USA) program packages. The applicability of the last approach for experimental IR-spectroscopic band assignment as well as for obtaining the stereo-structural information has been demonstrated in a series of organic systems and coordination complexes as heterocyclic (Ivanova 2005a, b), Cu(II) complexes (Ivanova and Mayer-Figge 2005), polymorphs (Ivanova 2005a, b; Koleva 2006), codeine derivatives (Kolev et al. 2006a, b) peptides and their Au(III) complexes, hydrochlorides and hydrogensquarates (Ivanova et al. 2006a, b; Kolev 2006; Kolev et al. 2006b, 2007). The theory of IR-LD spectroscopy and the employed polarized IR-spectra interpretation of difference-reduction procedure are given in more of the above citated references. The method consists of subtracting a perpendicular spectrum (IR_s), resulting at a 90° angle between the polarized light beam electric vector and the orientation of the sample, from a parallel one (IR_p), obtained with a co-linear mutual orientation. The recorded difference (IR_p-IR_s) spectrum divides the parallel (A_p) and perpendicular (A_s) integrated absorbencies of each band into positive contributions, originating from transition moments which form average angles with the orientation direction (**n**) between 0° and 54.7° and negative ones, corresponding to the transition moments between 54.7° and 90°. In the next operation, the perpendicular spectrum multiplied by the parameter ***c***, is subtracted from the parallel one and ***c*** varied till one or set of bands are eliminated. The simultaneous disappearance of bands in the resulting ($IR_p-\boldsymbol{c}IR_s$) reduced IR-LD spectrum indicates a co-linearity of the corresponding transition moments, thus giving rise to information regarding the mutual disposition of the molecular fragments and the application of the presented tool of the IR-spectroscopic pattern of (**I**) in solid state.

The FAB mass spectra were recorded on a Fisons VG autospect instrument employing 3-nitrobenzylalcohol as matrix. The elemental analysis was carried out according to the standard procedures for C, H (as CO_2, and H_2O) and N (by the Dumas method).

Quantum chemical calculations were performed with GAUSSIAN 98 program package (Frisch et al. 1998). The output fails are visualized by means of ChemCraft program (Zhurko and Zhurko 2005). The geometry of tyrosinamidium–hydrogenphosphate ions and water system was optimized at two levels of theory: second-order Moller–Pleset perturbation theory (MP2) and density functional theory (DFT) using 6-311++G** basis set. DFT method employed is B3LYP, which combines Becke's three-parameter non-local exchange functional with the correlation function of Lee, Yang and Parr0. Molecular geometry of the studied species was fully optimized by the force gradient method using Bernys' algorithm. For every structure, the stationary points found on the molecule potential energy hypersurfaces were characterized using standard analytical harmonic vibrational analysis. The absence of the imaginary frequencies, as well as that of negative eigenvalues of the second-derivative matrix, confirmed that the stationary points correspond to minima of the potential energy hypersurfaces. The calculation of vibrational frequencies and infrared intensities was checked for which kind of calculations performed agree best with the experimental data. The DFT method provides more accurate vibrational data, as far as the calculated

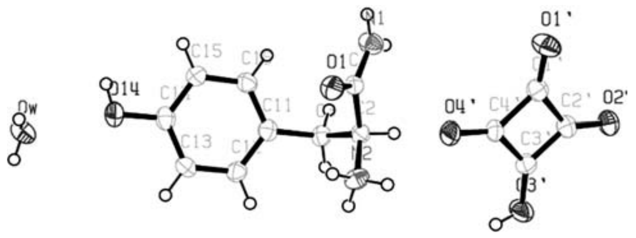


Fig. 1 ORTEP plot of tyrosinamide hydrogensquarate monohydrate

standard deviations are 10 cm^{-1} (B3LYP) and 19 cm^{-1} (MP2), respectively. So, the B3LYP/6-31G** data are presented for above discussed modes, where for the better correspondence between the experimental and theoretical values, a modification of the results using the empirical scaling factor 0.9614 is made. The vibrational analysis supported experimental IR-spectroscopic characterization. The normal modes of benzene ring are notes using Wilson notation (Varsanyi 1969). The presented theoretical approximation gives significantly different value when compared with the corresponding IR-spectroscopic frequencies in solid state for the so-called characteristic bands, where the participation of given group in intra or intermolecular interaction shifted the corresponding band. For last maxima, the deviation varies within 12–54 cm^{-1} for system studied.

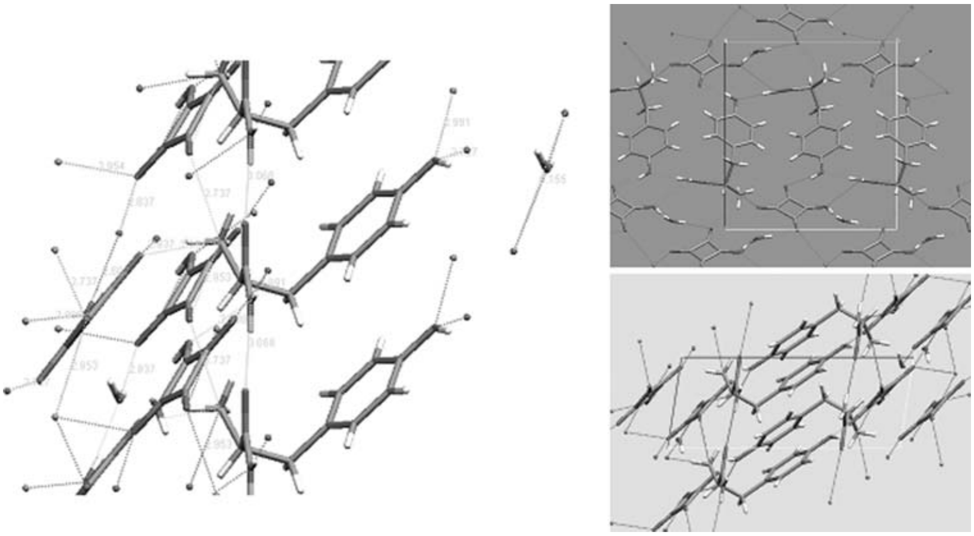
Result and discussion

Crystallographic data of (**I**), Fig. 1, show that the structure consists of 3D network of molecules joined by intermolecular $_{(Tyr)}OH\cdots O{=}C_{(Sq)}$ (2.727 Å), $O{=}C{-}NH_2\cdots OH_{(Tyr)}$ (2.991 Å), $O{=}C{-}NH_2\cdots OH_{(Sq)}$ (2.988 Å), $O{=}C{-}NH_2\cdots O{=}C{-}NH_2$ (3.068 Å), $N^+H_3\cdots O{=}C_{(Sq)}$ (2.737, 2.953, 2.954 Å), $OH_2\cdots O{=}C_{(Sq)}$ (2.839 Å) and $_{(Sq)}OH\cdots OH_2$ (2.607 Å) hydrogen bonds (Fig. 2). In the frame of the unit cell, the benzene rings planes of the tyrosinamide molecules close at an angle of 26.4(9)° (Scheme 2a), while the planes of the aromatic ring and amide fragments in one molecule closed at an angle of 46.4(4)° (Scheme 2b). The hydrogensquarate anions are flat with a deviation of total planarity of 1.9(8)°. According to the quantum chemical calculation, the interacting system tyrosinamide, hydrogensquarate ion and water molecule is characterized with the stabilization of the conformer of tyrosinamide E_{rel} of 3.4 kJ/mol (Scheme 3). Two types of strong intermolecular interaction are predicted theoretically, $N^+H_3\cdots O{=}C_{(Sq)}$ and $_{(Tyr)}OH\cdots OH_2$ with lengths of 2.508 and 2.656 Å, respectively. The N–H···O and O–H···O angles of 150.0(0)° and 150.6(8)° are obtained. The comparison between the theoretically predicted and experimentally observed values of bond lengths and angles shows a good correlation that resulted in a difference of less than 0.066 Å and 9.3(3)°, respectively.

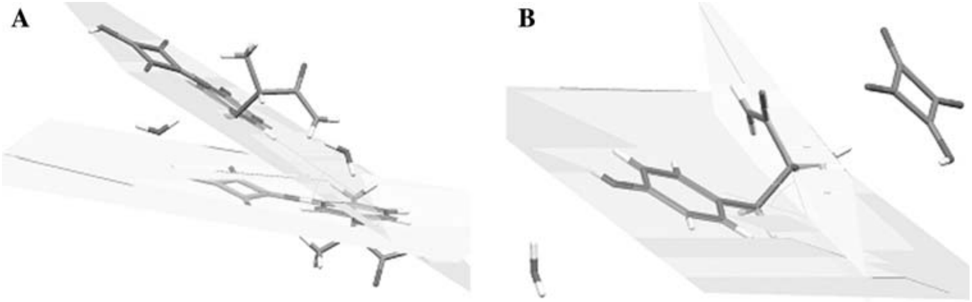
The dihedral angle values are reasonably predicted by B3LYP/6-31G** where the maximal deviation when compared with the crystallographic data do not differ by 4.7°, expecting the values of $CH_2{-}CH(N^+H_3){-}C = O$ and $NH_3{-}CH{-}C({=}O){-}NH_2$ to differ with 107.1° and 107.6°, respectively. These results are observed in the cases of peptide systems, where the intermolecular interactions in solid state resulted in a deviation of corresponding dihedral angle values, when compared with theoretical data of these angles connected with the functional groups included in the corresponding hydrogen bonding (Ivanova et al. 2006a, b; Kolev et al. 2006b, 2007; Kolev 2006).

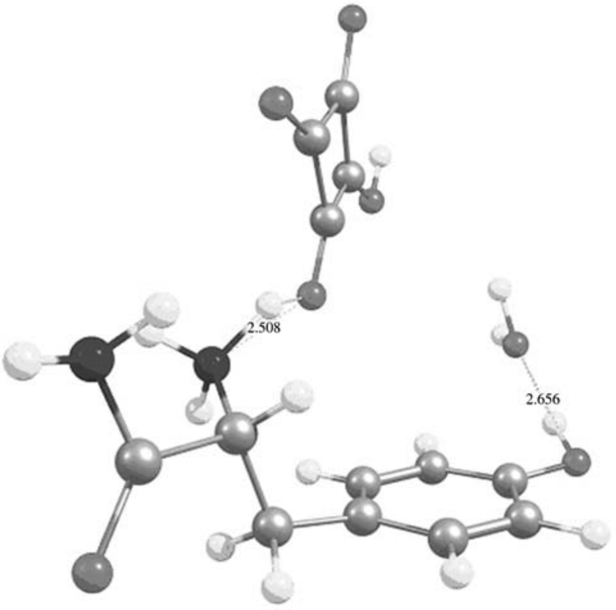
Non-polarized IR- and difference IR-LD spectra of (I) (Fig. 3) show that obtained macro-orientation of polycrystalline sample according to quantization supposed an

Fig. 2 Hydrogen bonding



Scheme 2 Unit cell of titled compound





Scheme 3 Optimized geometry of interacted system

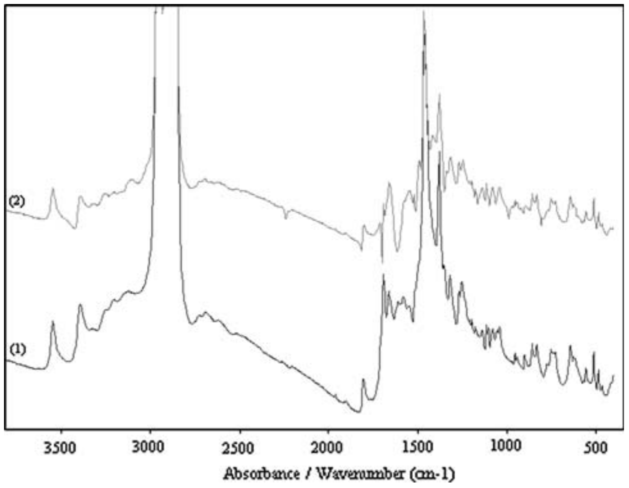


Fig. 3 Non-polarized IR-(*1*) and difference IR-LD (*2*) spectrum of tyrosinamide hydrogensquarate monohydrate

adequate interpretation of polarized data, that is, obtaining the structural information and IR-band assignment. The observed bands and their assignment are: 3,564 cm^{-1} ($\nu_{\mathrm{OH(solvent)}}$), 3,397 cm^{-1} ($\nu^{as}_{\mathrm{NH2}}$), 3,248 cm^{-1} ($\nu_{\mathrm{OH(Tyr)}}$, hydrogen bonded), 3,205 cm^{-1} (ν^{s}_{NH2}), 3,357–2,280 cm^{-1} region ($\nu^{as}_{\mathrm{N+H3}}$, $\nu^{as'}_{\mathrm{N+H3}}$, $\nu^{s}_{\mathrm{N+H3}}$), 1,801 cm^{-1} ($\nu^{s}_{\mathrm{C=O(Sq)}}$), 1,697 cm^{-1} ($\nu_{\mathrm{C=O}}$ (Amide I)), 1,709 cm^{-1} ($\nu^{as}_{\mathrm{C=O(Sq)}}$), 1,659 cm^{-1} ($\nu_{\mathrm{C=C(Sq)}}$), 1,667 cm^{-1} ($\delta^{as}_{\mathrm{N+H3}}$), 1,644 cm^{-1} ($\delta^{as'}_{\mathrm{N+H3}}$), 1,609 cm^{-1} (8a, benzene in-planemode), 1,580 cm^{-1} (8b, benzene in plane mode), 1,544 cm^{-1} ($\delta^{s}_{\mathrm{N+H3}}$), 1,517 cm^{-1} (19a, benzene in-plane mode), 857 cm^{-1} (11-γ_{CH}, benzene out-of-plane mode). This assignment is made using the linear-polarized IR-spectroscopic data and the so-called reducing difference procedure for the interpretation of the spectra of this type, which could be illustrated with following. The elimination of the band at 857 cm^{-1} (Fig. 4) leads to reduction of the maximum at 640 cm^{-1} due to near co-linearity of 11-γ_{CH} and $\gamma_{\mathrm{C=O(Sq)}}$ out-of-plane transition moments, which are disposed perpendicularly to the corresponding aromatic and hydrogensquarate skeletons.

In conclusion, the spectral and structural elucidation of a newly synthesized biologically active complex of L-tyrosinamide with squaric acid, whose structure is refined by single crystal X-ray diffraction method, is presented. The conformational behavior depending on the types of hydrogen bonding in solid phase is obtained comparing the crystallographic data with those of the theoretical DFT of interacting system tyrozinamidium cation, hydrogensquarate anion and a water molecule. Two types of intermolecular hydrogen bonds are predicted ($\mathrm{N^+H_3\cdots O_{(Sq)}}$ and $\mathrm{_{(Tyr)}OH...OH_2}$ with bond lengths of 2.508 and 2.656 Å, respectively) and a number of experimentally observed ones [$\mathrm{_{(Tyr)}OH\cdots O{=}C_{(Sq)}}$ (2.727 Å), $\mathrm{O{=}C{-}NH_2\cdots OH_{(Tyr)}}$ (2.991 Å), $\mathrm{O{=}C{-}NH_2\cdots OH_{(Sq)}}$ (2.988 Å), $\mathrm{O{=}C{-}NH_2\cdots O{=}C{-}NH_2}$ (3.068 Å), $\mathrm{N^+H_3\cdots O{=}C_{(Sq)}}$ (2.737, 2.953, 2.954 Å), $\mathrm{OH_2\cdots O{=}C_{(Sq)}}$ (2.839 Å) and $\mathrm{_{(Sq)}OH\cdots OH_2}$ (2.607 Å)] resulted in the observation that the difference between theoretical dihedral angle values and the experimental ones of $\mathrm{CH_2{-}CH(N^+H_3){-}C{=}O}$ and $\mathrm{NH_3{-}CH{-}C({=}O){-}NH_2}$ differed with 107.1° and 107.6°, respectively.

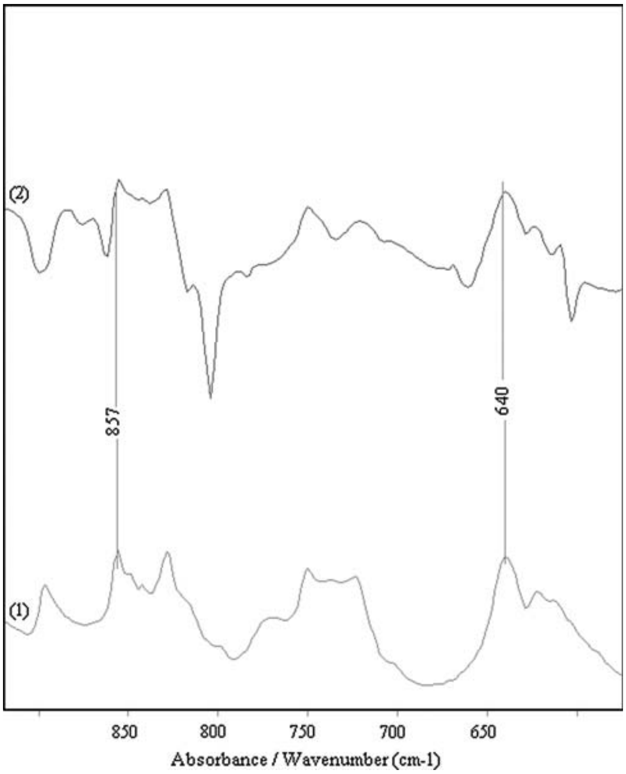


Fig. 4 Non-polarized IR-(*1*) and reduced IR-LD (*2*) spectrum of tyrosinamide hydrogensquarate monohydrate after elimination of the band at 857 cm^{-1}

The nature of the intermolecular hydrogen bonding affected as well on the IR-spectroscopic pattern of the system studied in solid state, where the deviation between predicted and observed bands assigned to so-called characteristic bands varies within 12–54 cm^{-1}. For other bands the applied theoretical approximation (B3LYP/6-31+G**) give a reasonable value because the difference between theoretical and experimental values is less then 9 cm^{-1}. As far as the presented new spectroscopic technique, applied in linear-polarized IR-spectroscopy in solid state, namely orientation of the samples as suspension in nematic liquid crystal gives not only experimental IR-assignment evidence, but also a structural information, the relationship structure-spectroscopic properties in vitro is performed. This knowledge is an important part for biological in vivo investigations using IR-spectroscopic tools, because every influence on the structure or the types of intermolecular interactions is directly connected with the changes on the IR-spectroscopic characteristics.

Supporting information

Crystallographic data for the structural analysis have been deposited with the Cambridge Crystallographic Data Centre, CCDC 631985. Copies of this information may be obtained from the Director, CCDC, 12 Union Road, Cambridge, CB2 1EZ, UK (Fax: +44 1223 336 033; e-mail: deposit@ccdc.cam.ac.uk or http://www.ccdc.cam.ac.uk).

Acknowledgments T.K. B.K. and M.S. wish to thank the DAAD for a grant within the priority program “Stability Pact South-Eastern Europe” and the Alexander von Humboldt Foundation.

References

Frisch MJ, Trucks GW, Schlegel HB, Scuseria GE, Robb MA, Cheeseman JR, Zakrzewski VG, Montgomery Jr JA, Stratmann RE, Burant JC, Dapprich S, Millam JM, Daniels AD, Kudin KN, Strain MC, Farkas Ö, Tomasi J, Barone V, Cossi M, Cammi R, Mennucci B, Pomelli C, Adamo C, Clifford S, Ochterski J, Petersson GA, Ayala PY, Cui Q, Morokuma K, Salvador P, Dannenberg JJ, Malick DK, Rabuck AD, Raghavachari K, Foresman JB, Cioslowski J, Ortiz JV, Baboul AG, Stefanov BB, Liu G, Liashenko A, Piskorz P, Komáromi I, Gomperts R, Martin RL, Fox DJ, Keith T, Al-Laham MA, Peng CY, Nanayakkara A, Challacombe M, Gill PMW, Johnson B, Chen W, Wong MW, Andres JL, Gonzalez C, Head-Gordon M, Replogle ES, Pople (1998) Gaussian JA 98, Gaussian, Inc., Pittsburgh, PA

Gilbert A, Antane M, Argentier T, Butera J, Francisco G, Freeden C, Gundersen E, Craceffa R, Herbst D, Hirth B, Lennox J, McFarlane G, Norton NW, Quaglito D, Sheldon J, Warga D, Wojdan A, Woods M (2000) Design and SAR of novel potassium channel openers targeted for urge urinary incontinence. Selective and potent benzulamino cyclobutenediones J Med Chem 43:1203–1214

Hruby M, Subr V, Kucka J, Kozempel J, Lebeda O, Sikora A (2005) Thermoresponsive polymers as promising new materials for local radiotherapy. Appl Radiat Isot 63:423–431

Ivanova BB (2005a) Solid state linear dichroic infrared spectral analysis of benzimidazoles and their N_1-protonated salts. Spectrochim Acta A 62A(1–3):58

Ivanova BB (2005b) Monoclinic and orthorhombic polymorphs of paracetamol–solid state linear dichroic infrared spectral analysis. J Mol Struct 738:233

Ivanova BB, Mayer-Figge H (2005) Crystal structure and Solid state IR-LD spectral analysis of new mononuclear Cu(II) complex with 4-aminopyridine. J Coord Chem 58:653

Ivanova BB, Arnaudov MG, Bontchev PR (2004) Linear-dichroic infrared spectral analysis of Cu(I)-homocysteine complex. Spectrochim Acta A 60A(4):855

Ivanova BB, Tsalev DL, Arnaudov MG (2006a) Validation of reducing-difference procedure for the interpretation of non-polarized infrared spectra of n-component solid mixtures. Talanta 69:822

Ivanova BB, Kolev T, Zareva SY (2006b) Solid-state IR-LD spectroscopic and theoretical analysis of glycine-containing peptides and their hydrochlorides. Biopolymers 82:587

Ivanova BB, Simeonov VD, Arnaudov MG, Tsalev DL (2007) Linear-dichroic infrared spectroscopy—validation and experimental design of the orientation technique as suspension in nematic liquid crystal”, Spectrochim Acta A 67(1):66–75

Kolev T (2006) Solid-state IR-LD spectroscopic and theoretical analysis of arginine-containing peptides. Biopolymers 83:39–45

Kolev T, Ivanova BB, Bakalska R (2006a) 6-O-Acetylcodeine and its hydrogensquarate - linear-dichroic infrared (IR-LD) spectroscopy. J Mol Struct 794:138

Kolev T, Zareva SY, Koleva BB, Spiteller M (2006b) Au(III) complexes of di- and tripeptide glycylglycine and glycylglycylglycine. Inorg Chim Acta 359:4367

Kolev T, Ivanova BB, Zareva SY (2007) Au(III)-complex with dipeptide glycyl-serine–linear polarized IR-spectroscopic, ^{1}H- and ^{13}C- magnetic resonance characterization. J Coord Chem 60:109

Koleva BB (2006) Polymorphs of aspirin—solid-state IR-LD spectroscopic and quantitative determination in solid mixtures. J Mol Struct 800:23

Kulathila R, Merkler KA, Merkler DJ (1999) Enzymatic formation of C-terminal amides. Nat Prod Rep 16:145

Marx KA, Lee JS, Sung C (2004) Electropolymerized films formed from the amphiphilic decyl esters of D- and L-tyrosine compared to *L*-tyrosine using the electrochemical quartz crystal microbalance. Biomacromolecules 5:1869

Merkler D (1994) C-terminal amidated peptides: production by the in vitro enzymatic amidation of glycine extended peptides and the importance of the amides for bioactivity. Enzymme Microb Technol 16:450

Onaran MB, Comeau AB, Seto CT (2005) Squaric acid-based peptidic inhibitors of matrix metalloprotease-1. J Org Chem 70:10792

Porter J, Archibald S, Child K, Critchley D, Head J, Linsley J, Parton T, Robinson M, Shock A, Taylor R, Warrellow G, Alexander R, Langham B (2000) Squaric acid derivatives as VLA-4 integrin antagonists. Bioorg Med Chem Lett 12:1051

Sheldrick GM (1995) SHELXTL, Release 5.03 for Siemens R3 crystallographic research system. Siemens Analytical X-Ray Instruments, Inc., Madison, USA

Sheldrick GM (1997) SHELXS97 and SHELXL97. University of Goettingen, Germany

Suwan S, Isobe M, Yamashita O, Minakata H, Imai K (1994) Silkworm diapause hormones structure-activity relationships indispensable role of C-terminal amides. Insect Biochem Mol Biol 24:1001

Sztaricskai F, Sum A, Roth F, Pelyvas I, Sandor S, Batta G, Herczegh P, Remenyi J, Miklan Z, Hudecz F (2005) A new class of semisynthetic anthracycline glycoside antibiotics incorporating a squaric acid moiety. J Antibiot 58:704

Tevyashova A, Sztaricskai F, Batta G, Herczegh P, Jeney A (2004) Formation of squarci acid amides of antracycline antibiotics. Synthesis and cytotoxic properties. Bioorg Med Chem Lett 14:4783

Varsanyi G (1969) Vibrational spectra of benzene derivatives. Academy Press, Budapest, pp 1–413

Vianini E, Palumbo M, Gatto B (2001) In vitro selection of DNA aptamers that bind L-tyrosinamide. Bioorg Med Chem 9:2543

Wilczynski KR, Wilson JW, Scott AS, Edison C, Haskell-Luevano C (2005) Structure-activity relationships of the unique and potent agouti-related protein (AGRP)-melanocortin chimeric Tyr-c[β-Asp-His-DPhe-Arg-Trp-Asn-Ala- Phe-Dpr]-Tyr-NH_2 peptide template. J Med Chem 48:3060

Xie J, Comeau AB, Seto CT (2004) quaric acid: a new motif for designing inhibitors of protein tyrosine phosphatases. Org Lett 6:83

Zhurko GA, Zhurko DA (2005) ChemCraft: tool for treatment of chemical data, Lite version build 08 (freeware)