Contents lists available at ScienceDirect

Solid State Nuclear Magnetic Resonance

An NMR crystallography study of the hemihydrate of 2′, 3′-O-isopropylidineguanosine

G.N. Manjunatha Reddy [a], Daniel S. Cook [b], Dinu Iuga [a], Richard I. Walton [b], Andrew Marsh [b], Steven P. Brown [a,*]

[a] Department of Physics, University of Warwick, Coventry CV4 7AL, UK
[b] Department of Chemistry, University of Warwick, Coventry CV4 7AL, UK

ARTICLE INFO

ABSTRACT

An NMR crystallography study of the hemihydrate of 2′, 3′-O-isopropylidineguanosine (Gace) is presented, together with powder X-ray diffraction and thermogravimetric analysis. ^{1}H double-quantum and ^{14}N–^{1}H HMQC spectra recorded at 850 MHz and 75 kHz MAS (using a JEOL 1 mm probe) are presented together with a ^{1}H–^{13}C refocused INEPT spectrum recorded at 500 MHz and 12.5 kHz MAS using eDUMBO-1$_{22}$ ^{1}H homonuclear decoupling. NMR chemical shieldings are calculated using the GIPAW (gauge-including projector augmented wave) method; good two-dimensional agreement between calculation and experiment is observed for ^{13}C and ^{1}H chemical shifts for directly bonded CH and CH$_3$ peaks. There are two Gace molecules in the asymmetric unit cell: differences in specific ^{1}H chemical shifts are rationalised in terms of the strength of CH-π and intermolecular hydrogen bonding interactions.

1. Introduction

The collaborative computational project for NMR crystallography (CCP-NC, www.ccpnc.ac.uk) defines NMR crystallography as the combined use of experimental NMR and computation to provide new insight, with atomic resolution, into structure, disorder and dynamics in the solid state. Building upon pioneering work, notably by Harris [1,2] and Emsley [3], NMR crystallography of solid-state structures adopted by organic molecules is becoming a vibrant research area. In particular, calculations of NMR parameters using the GIPAW [4–7] (gauge-including projector augmented wave) method have been performed for a wide variety of organic solids [8–35].

This paper considers 2′, 3′-O-isopropylidineguanosine; using the notation in Ref. [25], this is referred to as Gace (i.e., for guanosine acetonide). Such guanosine derivatives are of interest because of the rich supramolecular chemistry exhibited in organic solutions, on surfaces and in the solid state [36–38], with self assembly being characterised by ^{23}Na, ^{39}K and ^{87}Rb [39–43] as well as ^{1}H, ^{13}C, ^{14}N and ^{15}N [25,44,45] solid-state NMR.

In this work, the hemihydrate, Gace.0.5H$_2$O, for which the crystal structure has been solved by single crystal X-ray diffraction [46] is studied. A suite of two-dimensional high-resolution ^{1}H solid-state NMR experiments [47] that are applicable at natural isotopic abundance (^{1}H–^{13}C refocused INEPT [48], ^{1}H double-quantum (DQ) MAS [49], and ^{14}N–^{1}H HMQC [50–53] experiments) and that use J couplings or dipolar couplings to probe through-bond C–H connectivities and through-space H–H and N–H proximities are performed. In the NMR crystallography approach, experimental solid-state NMR spectroscopy (together with powder X-ray diffraction and thermogravimetric analysis) is complemented by GIPAW calculations of NMR chemical shifts.

* Corresponding author. Fax: +44 24 761 50897.
E-mail address: S.P.Brown@warwick.ac.uk (S.P. Brown).
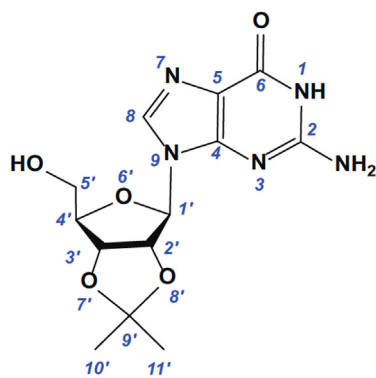
Gace

2. Experimental and computational details

Gace was purchased from Sigma-Aldrich (Gillingham, UK). PXRD data were collected at room temperature on a PANalytical X'Pert Pro MPD ($K\alpha_1$ $\lambda = 1.5406$ Å) equipped with monochromatic Cu $K\alpha_1$ radiation and a PIXcel detector from a sample held in an aluminium plate. Pawley fitting of the diffraction profile was used to obtain lattice parameters—TOPAS-Academic implemented with jEdit (version 4.3.1) was employed [54]. The goodness of fit is reported as the weighted (R_{wp}) and unweighted (R_p) profile parameters, defined in the conventional way [55].

Thermogravimetric analysis was performed using a Mettler Toledo DSC1-400 instrument. $\sim$10 mg of sample contained within a 40 μL aluminium sample pan was heated over the temperature range 25–200 °C at a constant heating rate of 10 °C per minute. A purge gas using helium was employed at a flow rate of 50 ml per minute.

Solid-state NMR experiments were performed using Bruker Avance III NMR spectrometers operating at a ^{1}H Larmor frequency of 500.1 MHz for ^{13}C–^{1}H experiments (^{13}C Larmor frequency of 125.8 MHz) or 850.2 MHz for ^{1}H and ^{14}N–^{1}H experiments (^{14}N Larmor frequency of 61.4 MHz). At 500 MHz, a Bruker 4 mm triple-resonance MAS probe (in double-resonance mode) was used, with 46 mg of Gace packed into the rotor, while at 850 MHz, a JEOL 1 mm double-resonance MAS probe was used, with 0.6 mg of Gace packed into the rotor. A recycle delay of 3 s (500 MHz) or 2 s (850 MHz) was used. In ^{13}C–^{1}H CP MAS and refocused INEPT experiments, SPINAL64 ^{1}H heteronuclear decoupling [56] with a pulse duration of 4.8 μs was applied for an acquisition time of 40 ms. The ^{1}H nutation frequency for pulses and decoupling was 100 kHz, except for ^{14}N–^{1}H experiments, where a ^{1}H 90° pulse duration of 2.0 μs was used.

^{13}C and ^{1}H chemical shifts are referenced with respect to neat TMS using adamantane as a secondary reference (38.5 ppm for the higher-ppm ^{13}C resonance [57] and 1.85 ppm for the ^{1}H resonance [58]). Experimental ^{13}C and ^{1}H chemical shifts are stated to an accuracy of $\pm$ 0.1 or 0.2 ppm, respectively. ^{14}N shifts were referenced to a saturated NH$_4$Cl aqueous solution at -352.9 ppm, corresponding to a primary reference of CH$_3$NO$_2$ at 0 ppm. To convert to the corresponding ^{15}N chemical shift scale frequently used in protein NMR, where the reference is liquid ammonia at -50 °C, it is necessary to add 379.5 to the given values [59].

Pulse sequences and coherence transfer pathway diagrams for the ^{1}H (SQ-DUMBO)–^{13}C SQ refocused INEPT [48], ^{1}H DQ MAS [49] using BABA (back-to-back) recoupling [60,61], and ^{14}N–^{1}H HMQC [51] two-dimensional experiments are shown in Fig. 5 of Ref. [48], Fig. 7 of Ref. [62], and Fig. 3a of Ref. [51]. For the ^{1}H (SQ-DUMBO)–^{13}C SQ refocused INEPT experiment, a 16-step phase cycle was used as described in Ref. [48]. For the ^{1}H DQ MAS experiment, a 16-step phase cycle was used to select $\Delta p = \pm 2$ on the DQ excitation pulses (4 steps) and $\Delta p = \pm 1$ (4 steps) on the z-filter 90° pulse, where p is the coherence order. For the ^{14}N–^{1}H HMQC experiment, a 4-step nested phase cycle was used to select changes in coherence order $\Delta p = \pm 1$ (on the first ^{1}H pulse, 2 steps) and $\Delta p = \pm 1$ (on the last ^{14}N pulse, 2 steps). A modified version of the pulse sequence was employed [25] whereby a second ^{1}H 90° pulse (90° out of phase with respect to the first 90° pulse) was applied immediately after the first ^{1}H 90° pulse and using phase inversion (0° and 180° every rotor period) [63] of the $n=2$ ($\nu_1 = 2\nu_R$) rotary-resonance recoupling (R^3) [64] pulses.

For the 2D ^{1}H–^{13}C refocused INEPT experiment, eDUMBO-1$_{22}$ homonuclear decoupling, [65,66] was employed during the ^{1}H evolution period and the τ and τ' spin-echo durations. The 32 μs eDUMBO-1$_{22}$ cycle was divided into 320 steps of 100 ns. Pulse sequences employing ^{1}H homonuclear decoupling use pre-pulses to take into account the tilting of the effective field away from the x and y plane of the rotation frame: [67] a pre-pulse duration of 0.8 μs was used. The scaling factor was determined experimentally (by using the ^{1}H chemical shifts of the resolved resonances in a ^{1}H 75 kHz MAS spectrum) as 1.60. For ^{13}C pulses, the ^{13}C 90° pulse length was 4.0 μs.

Calculations were performed using the CASTEP code [68], academic release version 6.1. Geometry optimisations and NMR chemical shift calculations using the GIPAW [4,5] method employed the PBE exchange correlation functional [69] with a semi empirical dispersion correction scheme due to Tkatchenko and Sheffler [70]—for more details of the implementation in NMR calculations see Ref. [33]. A plane wave basis set with ultra-soft pseudopotentials [71] with a maximum cut-off energy of 800 eV was used; integrals were taken over the Brillouin zone by using a Monkhorst–Pack grid of minimum sample spacing $0.08 \times 2\pi$ Å^{-1}. Geometry optimisation was performed with the unit cell parameters fixed, starting from the X-ray single-crystal structure [46] determined at ambient temperature of Gace.0.5H$_2$O, CSD code VUYMIL, $Z=4$ (166 atoms in the unit cell), space group $P2_1$, $Z'=2$, i.e., two distinct isopropylidineguanosine molecules and one water molecule in the asymmetric unit cell. The forces, energies and displacements were converged to better than 0.01 eV Å^{-1}, 0.0000004 eV, and 0.001 Å, respectively. Note that distances stated in this paper are extracted from this geometry optimised crystal structure.

3. Results

3.1. PXRD and TGA

Figs. 1 and 2 present powder X-ray diffraction (PXRD) and thermogravimetric analysis (TGA), respectively, for the Gace.0.5H$_2$O sample. In Fig. 1, satisfactory agreement following Pawley fitting of the diffraction pattern (this corresponds to refining only the unit cell parameters, i.e., there is no consideration of the atomic coordinates within the unit cell) with respect to the published single-crystal X-ray diffraction structure [46] is observed. Table 1 compares the refined unit cell parameters to those of the published structure, showing the good agreement. By TGA (see Fig. 2), a weight loss of 2.2% is observed. The loss of half a water molecule per formula unit corresponds to 9 g/mol, i.e., 2.7% of the molecular weight of 323.3 g/mol for Gace.0.5H$_2$O. Thus, the experimentally observed weight loss of 2.2% is close to, but not exactly the same as, that expected for Gace.0.5H$_2$O.