Journal of Fluorine Chemistry 129 (2008) 493–497

Contents lists available at ScienceDirect

Journal of Fluorine Chemistry

journal homepage: www.elsevier.com/locate/fluor

Crystal and molecular structure of 5-trifluorothymine, a metabolite from human urine: Role of fluorine in stacking and hydrogen bonded interactions

Manju Rajeswaran [a], Thamarapu Srikrishnan [b,*]

[a] Kodak Research Laboratories, Eastman Kodak Company, Rochester, NY 14650, USA
[b] Department of Cancer Biology, Roswell Park Cancer Institute, Buffalo, NY 14263, USA

ARTICLE INFO

Article history:
Received 4 March 2008
Accepted 5 March 2008
Available online 18 March 2008

Keywords:
Polyflourinated thymines
Stacking of fluorinated bases
Dimeric hydrogen bonding of nucleic acid bases

ABSTRACT

The crystal structure of the metabolite from urine, 5-trifluorothymine [5F_3T] has been determined by single crystal X-ray diffractometric methods. Crystals of 5F_3T are monoclinic, space group $P2_1/c$ with cell dimensions $a = 6.7468(2)$, $b = 15.0740(6)$, $c = 13.4405(6)$, $\beta = 90.412(2)$, $V = 1366.88(8)$, $Z = 8$ (two molecules per asymmetric unit). Crystal structure of 5F_3T was determined with 3039 independent data and refined by full-matrix least squares methods to a final reliability factor of 0.047. Molecules of 5F_3T are connected by dimeric type of N–H···O hydrogen bonding linking molecules related by a center of inversion into an extensive layer of dimeric molecules. These layers are stacked on top of each other at a stacking distance of 3.280 Å with a head-to-head stacking of the fluorine atoms on top of each other with no hydrogen bonding involving the fluorine atoms.

1. Introduction

Certain purine and pyrimidine analogs readily replace the natural bases in nucleic acids if they are present during replication. Halogenated nucleic acids have been known for well over 30 years since it was first pointed out that 5-bromouracil and 5-iodouracil could be incorporated into the nucleic acids of *Streptococcus faecalis* [1] and they inhibit the growth of *S. faecalis*. Later it was shown that when *E. coli* cells are grown in the presence of 5-bromouracil, the latter is incorporated into the DNA of *E. coli* [2]. This provided the impetus for the synthesis of several 5-fluoro nucleic acid bases and their use as chemotherapeutic agents. In our ongoing studies of several of these analogs, we have synthesized and investigated the structure and properties of 5-fluorocytidine [3] and 5-fluorouridine [4]. The pyrimidine antimetabolite Ftorafur [FT; 5-fluoro-1-(tetrahydro-2-furyl)uracil] has shown significant antitumor activity in several adenocarcinomas with a spectrum of chemotherapeutic activity similar to, but less toxic than, 5-fluorouracil (5-FU) [5–7]. It is considered as a prodrug that acts as a depot form of 5-FU, and hence the two drugs exhibit a similar spectrum of chemotherapeutic activity [8–10]. Hydroxyl derivatives: 2′-hydroxyftorafur (III), 3′-hydroxyftorafur (IV) and 2′,3′-dihydroxyftorafur (II) were synthesized and X-ray and NMR studies of these hydroxyl derivatives have been investigated [11] in our laboratories to study the structural and conformational features of Ftorafur and its metabolites in the solid and solution states. One interesting feature in the chemistry of pyrimidines is the susceptibility of the 5,6-double bond to 1,4-nucleophilic addition reactions. Several pyrimidine derivatives have proven to be active antitumor, antipyretic and anti-inflammatory agents. The basic rationale in the design of these fluorinated analogs is the fact that fluorine does not influence the molecular geometry of the molecule but can change substantially the biological activity due to the alterations in the electronic and solubility properties. Polyfluorinated analogs such as trifluorothymine [F_3T], 5-trifluoromethyl-5,6-dihydrouracil [DHF_3T] and 5-trifluoromethyl-2′-deoxyuridine [F_3TdR] have been detected as metabolites in human urine [12]. A methodology to synthesize several *N*-polyfluorinated alkyl derivatives of uracil and 5-substituted uracil and X-ray structure of 1-mono (1H, 1H, 2H, 2H-perfluorooctyl)-5-trifluoromethyluracil has been published in an earlier issue of this journal [13]. In this paper, we describe the crystal structural features of F_3T.

2. Results and discussion

The isolation, characterization and study of the structure of metabolites in human urine is a very important field of biochemical and biophysical research and is a necessary prelude to understanding the biochemical metabolic processes in the disease process. In our institute, we have successfully isolated, purified and characterized well over forty purine and pyrimidine derivatives that are excreted in human urine [14–16]. Some

* Corresponding author. Tel.: +1 716 845 8302; fax: +1 716 845 4928.
E-mail address: thamarapu.srikrishnan@roswellpark.org (T. Srikrishnan).

0022-1139/$ – see front matter © 2008 Elsevier B.V. All rights reserved.
doi:10.1016/j.jfluchem.2008.03.003

urinary nucleosides such as 5-carbamoylmethyluridine [ncm(5)u], 5-carboylmethyl-2-thiouridine [nmc(5) s(2)u], N^2-dimethylguanine, 1-methylinosine and *N*-purin-6-ylcarbamoyl-L-threonine (t^6A) are derived from a turnover of t-RNA, whereas others such as N^6-succinyladenosine, orotidine, orotoic acid and 5-aminoimdiazole-4-carboxamide riboside result from the metabolism of purine and pyrimidine anabolic intermediates. In the urine of colon carcinoma patients, the t-RNA derived metabolites are elevated compared to that found in normal humans. Elevated levels of N^2-dimethylguanine have been observed in breast carcinoma patients and in chronic myelogenic leukemia [17,18] and elevated levels of N^6-succinyladenosine are found in urines of patients with prostate and liver adenocarcinoma [19]. As part of an ongoing program on the structures of nucleosides and related substances present in urine of cancer patients, we have carried out crystal structural investigations of several of these compounds [20–24]. These studies are extremely useful for assessing the tumor burden as well as for determining their therapeutic effectiveness.

A number of fluorinated metabolites are observed in urine, whole livers and blood samples. Polyfluorinated analogs such as [F_3T], [DHF_3T] and [F_3TdR] have been detected as metabolites in human urine [25] which were characterized by preparing authentic samples of F_3T prepared synthetically [26]. 5-Trifluoromethyluracil is synthesized starting with trifluoroacetone, which is converted into the cyanohydrin and then into cyanohydrin acetate which was pyrolyzed to trifluoroacrylonitrile. To trifluoroacrylonitrile, hydrogen bormide in methanol was added and the resulting product β-bromo-α-trifluoromethylpropionamide was formed. This product was condensed with urea or *N*-acetylurea to yield α-trifluoromethyl-β-ureido propionamide or its acetyl derivative. The ureidoamides were cyclized to 5-trifluoromethyl-5,6-dihydrouracil by reflexing in hydrochloric acid which was enzymatically converted into 5-trifluoromethyluracil.

Table 1
Crystal data and structure refinement for 5-trifluorothymine [5F_3T]

Compound name	5-Trifluorothymine
CCDC deposit no.	CCDC-668332
Color/shape	Colorless/rectangular
Chemical formula	$C_5H_3F_3N_2O_2$
Formula weight	180.09
Temperature	293(2) K
Crystal system	Monoclinic
Space group	$P2_1/c$
Unit cell dimensions	a = 6.7468(2) Å b = 15.0740(6) Å c = 13.4405(6) Å α = 90.0° β = 90.412(2)° γ = 90.0°
Volume (Å^3)	1366.88(8)
Z	8 (two per asymmetric unit)
Density observed (mg/m^3)	1.74
Density calculated (mg/m^3)	1.750
Absorption coefficient (mm^{-1})	0.186
Diffractometer	Kappa CCD
Radiation/wavelength	Mo Kα/0.71073 Å
Crystal size	0.50 mm × 0.20 mm × 0.10 mm
θ Range for data collection	3.32–27.34°
Index ranges	$-8 \le h \le 8$; $-19 \le k \le 19$; $-17 \le l \le 16$
Independent/observed reflections	3039 ($I \le 3\sigma$)
Refinement method	Full-matrix least squares on F^2
Computing	SHELXTL (Brucker 2000)
Data/restraints/parameters	3039/0/217
Goodness of fit on F^2	1.031
Function minimized	$\sum[\lvert F_o^2\rvert - (1/k)\lvert F_c^2\rvert]$
Final R indices [$I > 2\sigma(I)$]	R_1 = 0.0468, wR_2 = 0.1166
R indices (all data)	R_1 = 0.0738, wR_2 = 0.1306
Large diff. peaks and hole	0.317 and −0.238

Table 2
Final fractional positional parameters and their estimated standard deviations

Atom	x/a	y/b	z/c	B (Å^2)
Molecule A				
F(1)	5925(3)	3958(1)	4890(1)	93(1)
F(2)	5475(3)	2819(1)	5788(1)	83(1)
F(3)	2987(2)	3523(1)	5223(1)	90(1)
O(2)	4579(2)	5645(1)	9026(1)	52(1)
O(4)	8262(2)	4175(1)	6765(1)	67(1)
N(1)	2982(2)	4988(1)	7730(1)	41(1)
N(3)	6372(2)	4907(1)	7871(1)	44(1)
C(2)	4635(3)	5209(1)	8252(2)	39(1)
C(4)	6616(3)	4402(1)	7019(2)	45(1)
C(5)	4758(3)	4193(1)	6509(2)	43(1)
C(6)	3037(3)	4493(1)	6884(2)	43(1)
C(7)	4791(4)	3632(2)	5607(2)	58(1)
Molecule B				
F(1B)	1258(3)	2484(1)	7067(1)	79(1)
F(2B)	1302(3)	1423(1)	8099(1)	98(1)
F(3B)	−1444(2)	1957(1)	7586(1)	84(1)
O(2B)	286(2)	4482(1)	11058(1)	42(1)
O(4B)	3895(2)	3044(1)	8730(1)	51(1)
N(1B)	−1340(2)	3596(1)	9962(1)	41(1)
N(3B)	2046(2)	3727(1)	9900(1)	39(1)
C(2B)	325(3)	3961(1)	10350(1)	36(1)
C(4B)	2256(3)	3171(1)	9077(1)	39(1)
C(5B)	393(3)	2811(1)	8720(2)	41(1)
C(6B)	−1308(3)	3033(1)	9171(2)	44(1)
C(7B)	396(3)	2173(2)	7870(2)	53(1)

U(eq) is defined as one third of the trace of the orthogonalized Uij tensor.

A survey of the Cambridge Crystallographic data bank revealed very few structures with ployfluorinated purines and pyrimidines. The crystal structure of F_3T was undertaken to study the structure of this metabolite in the solid state as well as to evaluate the role played by the polyfluorinated ions in the molecular packing and hydrogen bonding. The compound crystallizes in the space group $P2_1/c$. The crystal and structure refinement data are given in Table 1. Table 2 gives the final atomic coordinates in the two

Table 3
Final bond distances (in Å) and bond angles (°) in 5-trifluorothymine

Bond distance	Molecule A	Bond distance	Molecule B
F(1)–C(7)	1.329(3)	F(1B)–C(7B)	1.317(3)
F(2)–C(7)	1.331(3)	F(2B)–C(7B)	1.319(3)
F(3)–C(7)	1.328(3)	F(3B)–C(7B)	1.336(3)
O(2)–C(2)	1.232(2)	O(2B)–C(2B)	1.234(2)
O(4)–C(4)	1.213(2)	O(4B)–C(4B)	1.219(2)
N(1)–C(2)	1.355(2)	N(1B)–C(2B)	1.352(2)
N(1)–C(6)	1.359(3)	N(1B)–C(6B)	1.361(3)
Bond angle	**Molecule A**	**Bond angle**	**Molecule B**
C(2)–N(1)–C(6)	122.72(16)	C(2B)–N(1B)–C(6B)	122.49(16)
C(2)–N(3)–C(4)	127.04(17)	C(2B)–N(3B)–C(4B)	126.88(16)
O(2)–C(2)–N(1)	122.61(17)	O(2B)–C(2B)–N(1B)	122.26(17)
O(2)–C(2)–N(3)	121.88(17)	O(2B)–C(2B)–N(3B)	122.09(16)
N(1)–C(2)–N(3)	115.50(17)	N(1B)–C(2B)–N(3B)	115.65(17)
O(4)–C(4)–N(3)	120.09(19)	O(4B)–C(4B)–N(3B)	119.71(18)
O(4)–C(4)–C(5)	126.3(2)	O(4B)–C(4B)–C(5B)	127.00(19)
N(3)–C(4)–C(5)	113.58(17)	N(3B)–C(4B)–C(5B)	113.29(16)
C(6)–C(5)–C(4)	119.45(19)	C(6B)–C(5B)–C(4B)	119.93(18)
C(6)–C(5)–C(7)	121.13(19)	C(6B)–C(5B)–C(7B)	120.73(19)
C(4)–C(5)–C(7)	119.39(18)	C(4B)–C(5B)–C(7B)	119.31(18)
C(5)–C(6)–N(1)	121.71(18)	C(5B)–C(6B)–N(1B)	121.74(18)
F(3)–C(7)–F(1)	107.1(2)	F(1B)–C(7B)–F(2B)	106.8(2)
F(3)–C(7)–F(2)	105.8(2)	F(1B)–C(7B)–F(3B)	105.5(2)
F(1)–C(7)–F(2)	105.8(2)	F(2B)–C(7B)–F(3B)	106.6(2)
F(3)–C(7)–C(5)	111.69(19)	F(1B)–C(7B)–C(5B)	113.6(2)
F(1)–C(7)–C(5)	113.3(2)	F(2B)–C(7B)–C(5B)	112.1(2)
F(2)–C(7)–C(5)	112.6(2)	F(3B)–C(7B)–C(5B)	111.65(18)

Table 4
Hydrogen bond distances (Å) and angles (°)

No.	Donor (D)	Hydrogen (H)	Acceptor (A)	Distances in Å (D–H)	Angle (°)			Symmetry
					H···A	D···A	D–H···A	
1	N1	H1	O2B	0.860	2.049	2.865	158.3	[−x, −y + 1, −z + 2]
2	N3	H3	O2B	0.860	1.967	2.822	172.4	[−x + 1, −y + 1, −z + 2]
3	N1B	H1B	O2	0.860	1.994	2.824	161.9	[−x, −y + 1, −z + 2]
4	N3B	H3B	O2	0.860	2.000	2.849	169.0	[−x + 1, −y + 1, −z + 2]
5	C6	H6	O4	0.93	2.46	3.260	144.0	[x − 1, y, z]
6	C6B	H6B	O4B	0.93	2.48	3.286	145.0	[−x − 1, −y − z]

molecules in the asymmetric unit. Table 3 gives the final bond distances and angles in the two molecules. The hydrogen bond distances and angles are given in Table 4. Fig. 1 gives an ORTEP diagram of the two molecules. Fig. 2 gives the hydrogen-bonding scheme linking the molecules into a dimeric network of head-to-tail layer. These layers are packed with a stacking distance of 3.280 Å. The fluorine atoms are stacked into a head-to-head arrangement with no hydrogen bonding involving the fluorine atoms (Fig. 3).

The crystal structure of thymine has been well characterized [27,28]. It crystallizes in the space group $P2_1/c$ and the crystal structure consists of planar thymine molecules linked together by N–H···O hydrogen bonding [2.810–2.836 Å] with the molecules lined up in a head-to-head fashion. When the methyl group is fluorinated, as in our structure, the molecules are interconnected by dimeric N–H···O hydrogen bonds involving N1, N3 and the keto oxygen O2. O4 of both the molecules are also involved in a long C–H···O hydrogen bonding involving the C6 of symmetry related molecule (see Table 4). The fluorine atoms are stacked into a head-to-head arrangement with no hydrogen bonding involving the fluorine atoms. In the trigonal form of trifluorinated thymine, a complicated infinite array is formed with the uracil molecules hydrogen bonded in both a head-to-tail and side-by side fashion [29]. The CF_3 groups of the thymine molecule form a pseudo-hexagonal channel and their orientation is stabilized by a rather long C–H···F interactions [30,31]. In the crystal structure of 5-fluorouracil [32], the molecules are hydrogen bonded in a ring around the pseudo fourfold axis within each layer, with several fluorine atoms localized in the same area contributing to a high electronegative concentration. Molecular adducts of 5-fluorouracil with cytosine and with thymine have been studied by Voet and Rich [33] about 40 years ago and more recently by Portalone and Colapietro [34] and Barnett et al. [35] with high precision data. In the hydrated crystal structure of 1:1 complex of cytosine and 5-fluorouracil [Cytfur] the molecules form a layered structure connected by hydrogen bonds with the disordered water molecules running through these channels. In the crystalline complex of 5-fluorouracil and thymine, the crystal structure comprises of interpenetrating hydrogen bonded nets, with four independent hydrogen bonds.

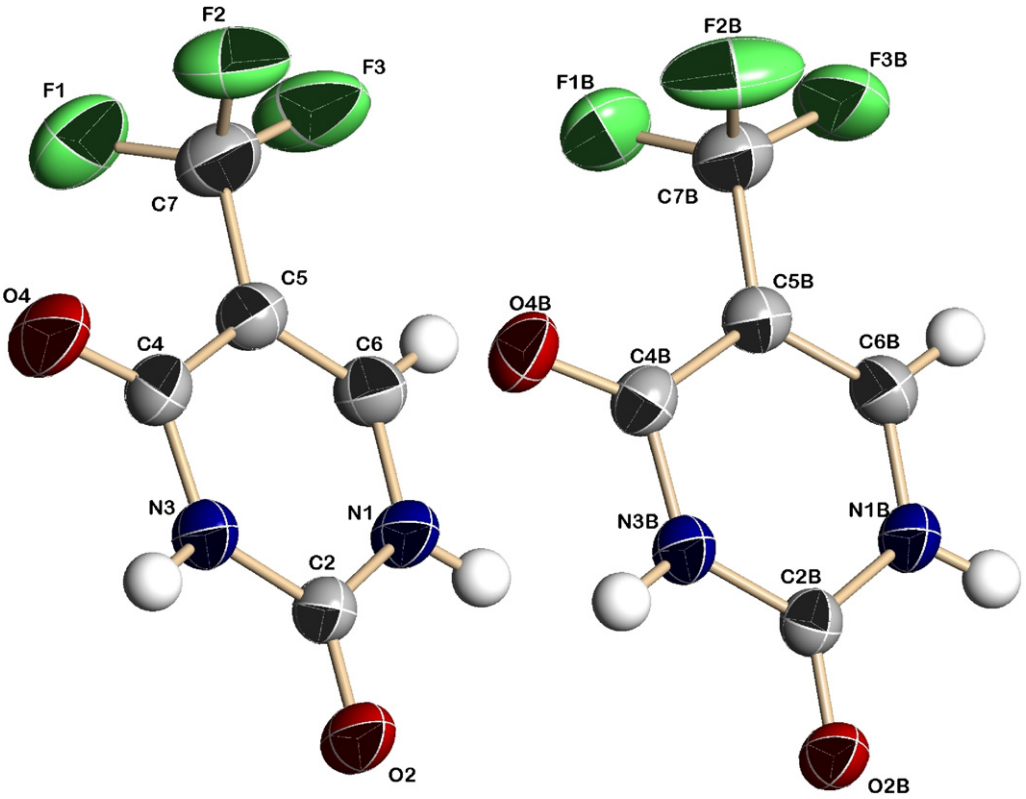

Fig. 1. ORTEP diagram of the two molecules of F_3T in the asymmetric unit.

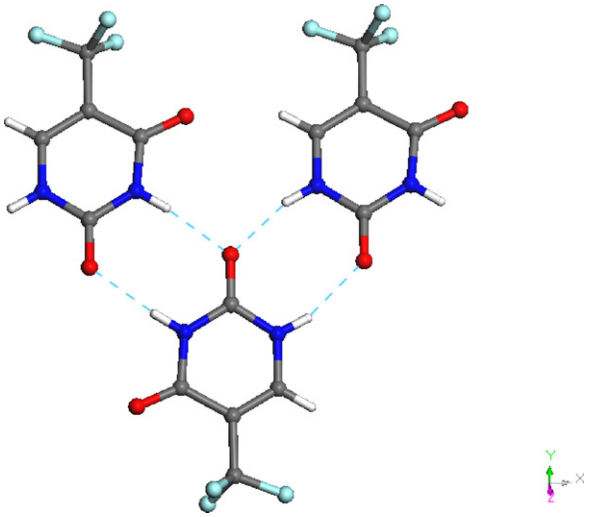

Fig. 2. Diagram illustrating the hydrogen scheme between the molecules in the unit. The molecules are connected by a network of N–H···O hydrogen bonds.

It is noteworthy to point that in the crystal structures of several fluorinated pyrimidines carried out in our laboratory such as 5-fluorocytidine [3], 5-fluorouridine [4] or 2′-hydroxyftorafur [11], 3′-hydroxyftorafur [11] or 2′,3′-dihydroxyftorafur [11], the fluorine atom does not take in the hydrogen bonding scheme. The presence of the highly electronegative fluorine atoms gives rise to F···F intermolecular interactions, which also extend into a 3D network. The two shortest F···F intermolecular distances are 3.066 and 3.495 Å respectively which is slightly larger than the value of 2.821(6) Å observed in the polyfluorinated structure of 1-mono (1H, 1H, 2H, 2H-perfluorooctyl)-5-trifluoromethyluracil [13].

The excretion, distribution and metabolism of $5F_3T$ and 5-trifluoromethyl-2′-deoxyuridine have been very well investigated by Heldelberger et al. [25] and Chheda [14]. The ring of these compounds is not metabolically degraded. The nucleoside is cleaved to the pyrimidine base and 5-carboxyuracil is the only catabolite formed. This is in sharp contrast to 5FU and FUDR where complete degradation of the pyrimidine ring takes place [36–40]. The degradation was shown to take place through the following sequence of reactions: 5FUDR → FU → dihydro-FU → α-fluoro-β-ureidopropionic acid → α-fluoro-β-guanidopropionic acid → urea or carbon dioxide and ammonia and α-fluoro-β-alanine.

3. Experimental

Excellent rectangular crystals of the compound were obtained by a slow evaporation of an aqueous solution of the compound. Crystals of $5F_3T$ ($C_5H_3F_3N_2O_2$) are monoclinic, space group $P2_1/c$, with cell dimensions $a = 6.7468(2)$, $b = 15.0740(2)$, $c = 13.4405(2)$ Å, $\beta = 90.412(2)^\circ$, $V = 1366.88(8)$ Å^3, $Z = 8$ (two molecules in the asymmetric unit). A crystal of approximate dimensions 0.5 mm × 0.2 mm × 0.1 mm was chosen for data collection. Diffraction data were collected at 298 K using a Nonius Kappa-CCD diffractometer with graphite-monochromated Mo Kα radiation ($\lambda = 0.71073$ Å). A total of 194 frames were collected using phi plus omega scans to fill the asymmetric unit with a scan range of 2° and a counting time of 200 s/°. The first ten frames were used for indexing reflections using the DENZO [41] package and refined to obtain final cell parameters. Table 1 gives the crystal data and the parameters used in the refinement of the structure of $5F_3T$. Data reductions were performed using DENZO-SMN [41]. The structure was solved by direct methods and refined by full-matrix least squares on F^2 with anisotropic displacement parameters for the non-hydrogen atoms using Bruker, SHELXTL [42] version 6.10. Hydrogen atoms were included in idealized positions. The

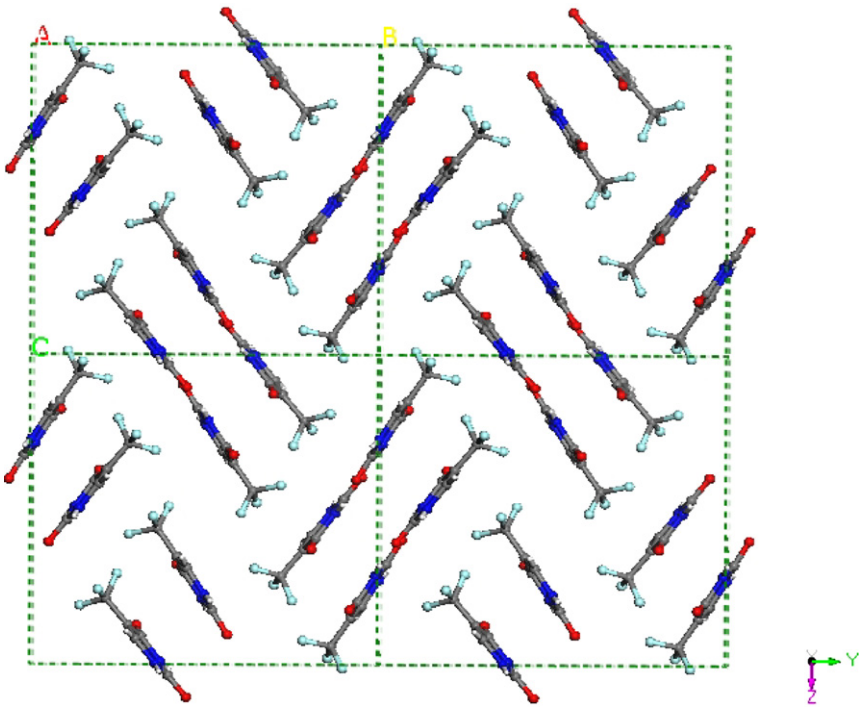

Fig. 3. Packing diagram illustrating the stacking interactions. The molecules linked by dimeric hydrogen bonding are stacked in a head-to-tail fashion in the unit cell forming a layer-like arrangement. These layers are stacked (stacking distance 3.280 Å) in a head-to-head manner with the fluorine atoms stacked on top of each other. The fluorine atoms do not take part in any hydrogen-bonded interactions but contribute to some non-bonded interactions.

structure refined to a goodness of fit (GOF)[1] of 1.031 and final residuals[2] of $R_1 = 0.0468\%$ ($I > 2\sigma(I)$), $wR_2 = 0.0738\%$ ($I > 2\sigma(I)$). A total of 3039 reflections were employed for 217 parameters determination, resulting in a data-to-parameter ratio of ~14. The final fractional coordinates, equivalent isotropic displacement parameters [U(eq)] of the atoms in the structure, bond lengths and angles and torsion angles are deposited with the manuscript.

Acknowledgements

This investigation was supported by Grant NIH-GM 24864 and the New York State department of health. We thank the referees and the editor for their very constructive suggestions and comments which were incorporated in the revised manuscript.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.jfluchem.2008.03.003.

References

[1] D.B. Dunn, J.D. Smith, S. Zamenhof, G. Griboff, Nature 174 (1954) 305–306.
[2] S. Zamenhof, G. Griboff, N. Marullo, Biochim. Biophys. Acta 13 (1954) 459–470.
[3] S.D. Soni, T. Srikrishnan, J.L. Alderfer, Nucleosides Nucleotides 15 (1996) 1945–1957.
[4] S.D. Soni, T. Srikrishnan, Nucleosides Nucleotides Nucleic Acids 23 (2004) 1779–1795.
[5] N.G. Blokhina, E.K. Vozny, A.M. Garin, Cancer 30 (1972) 390–392.
[6] N.I. Karev, N.G. Blokhina, E.K. Vozhy, M.P. Pershin, Neoplasma 19 (1972) 347–350.
[7] N. Valdidieso, G.P. Bodey, J.A. Gottlieb, E.J. Freirich, Cancer Res. 36 (1976) 1821–1824.
[8] A.M. Cohen, Drug Metab. Dispos. 3 (1975) 303–308.
[9] S. Fujimoto, T. Akao, B. Itoh, I. Koshizuka, K. Koyano, Y. Kitusakawa, M. Takahashi, T. Minami, H. Ishigami, Y. Nomura, K. Itoh, Cancer Res. 36 (1976) 33–36.
[10] A.Z. Smolyanaskaya, O.A. Tugarinov, Neoplasma 19 (1972) 341–345.
[11] D.M. Stokes, B. Paul, J.L. Alderfer, R.M. Wollman, T. Srikrishnan, Nucleosides Nucleotides Nucleic Acids 21 (2002) 863–890.
[12] M. Tandon, P. Kumar, G. Wiebe, L.I. Wiebe, Biochem. Pharm. 44 (1992) 2223–2228.
[13] O.D. Gupta, B. Twamley, R.L. Kirchmeier, J.M. Shreeve, J. Flourine Chem. 106 (2000) 199–204.
[14] G.B. Chheda, in: G. Fasman (Ed.), 3rd ed., Handbook of Biochemistry, vol. 1, Chemical Rubber Company, Cleveland, OH, 1975, pp. 251–270.
[15] G.B. Chheda, S.P. Dutta, A. Mittleman, J.A. Montgomery, S.K. Sethi, J.A. McCloskey, H.B. Patrzyc, Cancer Res. 45 (1985) 5958–5963.
[16] L. Pendyala, J.W. Cowens, G.B. Chheda, S.P. Dutta, P.J. Creaven, Cancer Res. 48 (1988) 3533–3536.
[17] D. Heldman, M.R. Graver, C.F. Speicher, R.W. Twewyn, J. Lab. Clin. Med. 101 (1983) 783–792.
[18] D.C. Tomey, T.P. Waakas, D. Altmann, C.W. Gehrke, R.W. Zumalt, J. Snyder. Cancer 35 (1975) 1095–1100.
[19] S.P. Dutta, A. Grossberg, A.K. Bhargava, G.B. Chheda, Anticancer Res. 6 (1986) 135–138.
[20] R. Parthasarathy, J.M. Ohrt, G.B. Chheda, Biochemistry 16 (1977) 4999–5008.
[21] A.V. Arakali, J. McCloskey, R. Parthasarathy, J.L. Alderfer, G.B. Chheda, Nucleosides Nucleotides Nucleic Acids 16 (1997) 2193–2218.
[22] T. Srikrishnan, R. Parthasarathy, W. Pfleiderer, N.C. De, G.B. Chheda, Nucleosides Nucleotides 16 (1997) 239–255.
[23] T. Srikrishnan, R. Parthasarathy, J.L. Alderfer, S.P. Dutta, G.B. Chheda, Nucleosides Nucleotides 7 (1988) 45–60.
[24] T. Srikrishnan, R. Parthasarathy, N.C. De, G.B. Chheda, Acta Cryst. C39 (1983) 1441–1445.
[25] C. Heldelberger, J. Bohar, B. Kampschroer, Cancer Res. 25 (1965) 377–381.
[26] C. Heidelberger, D.G. Parsons, D.C. Remy, J. Med. Chem. 7 (1964) 1–5.
[27] K. Ozeki, N. Sakabe, J. Tanaka, Acta Cryst. B25 (1969) 1038–1045.
[28] G. Portoalone, L. Bencivenni, M. Colapietro, A. Pieretti, F. Ramondo, Acta Chem. Scand. 53 (1999) 57–59.
[29] B. Twamley, O.D. Gupta, J.M. Shreeve, Acta Cryst. E58 (2002), o1040–o1042.
[30] G.R. Desiraju, T. Steiner. The Weak Hydrogen Bond in Structural Chemistry & Biology, IUCr Monograph No. 9, Oxford Press.
[31] L.J. Prins, D.N. Reinhoudt, P. Timmerman, Angew. Chem. (Int. Ed.) 40 (2001) 2382–2426.
[32] L. Fallon III, Acta Cryst. B29 (1973) 2549–2556.
[33] D. Voet, A. Rich, J. Am. Chem. Soc. 91 (1969) 3069–3075.
[34] G. Portalone, M. Colapietro, Acta Cryst. C63 (2007), o423–o425.
[35] S.A. Barnett, A.T. Hulme, D.A. Tocher, Acta Cryst. C62 (2006), o412–o415.
[36] N.K. Chaudhuri, B.J. Montag, C. Heidelberger, Cancer Res. 18 (1958) 318–328.
[37] N.K. Chaudhuri, K.L. Mukherjee, C. Heidelberger, Biochem. Pham. 1 (1959) 328–341.
[38] E. Harbers, N.K. Chaudhuri, C. Heidelberger, J. Biol. Chem. 234 (1959) 1255–1262.
[39] K.L. Mukherjee, J. Bohar, D. Wentland, F.J. Ansfield, C. Heidelberger, Cancer Res. 23 (1963) 49–66.
[40] K.L. Mukherjee, C. Heidelberger, J. Biol. Chem. 235 (1960) 433–437.
[41] Z. Otwinowski, W. Minor, in: C.W. Carter Jr., R.M. Sweets (Eds.), Methods in Enzymol., 276: Macromolecular Crystallography, Part A, Academic Press, pp. 307–326.
[42] SHELXTL: Structure Analysis Program, Version 6.10, Bruker Analytical X-ray Systems, Madison, WI, 2000.

[1] GOF $= [\sum[w(F_o^2 - F_c^2)^2]/(n-p)]^{1/2}$, where n and p denote the number of data and parameters.

[2] $R_1 = (\sum||F_o| - |F_c||)/\sum|F_o|$; $wR_2 = [\sum[w(F_o^2 - F_c^2)^2]/\sum[w(F_o^2)^2]]^{1/2}$, where $w = 1/[\sigma^2(F_o^2) + (a \cdot P)^2 + b \cdot P]$ and $P = [(\mathrm{Max}; 0, F_o^2) + 2 \cdot F_c^2]/3$.

Crystallographic data for this structure has been deposited with the Cambridge Crystallographic Data Centre as supplementary data no. CCDC 668332. Copies of the data may be obtained free of charge upon request from The Director, Cambridge Crystallographic Data Centre, 12 Union Road, Cambridge CB2 1EZ, UK (fax: +44 1223 336033; e-mail: deposit@ccdc.cam.ac.uk; WEB: http://www.csdc.cam.ac.uk).