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X-ray crystal structure of N'-[(1E)-1-(2,4-dihydroxyphenyl)-ethylidene]pyridine-4-carbohydrazide

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Abstract: Schiff's base of isonicotinyl hydrazide with 2',4'-dihydroxy acetophenone (INH-RA) has been designed and synthesized as a part of library enumeration targeting the NS2B-NS3 protease of Dengue virus. Slow evaporation from methanol results in the formation of monoclinic crystals C2/c space group with eight molecules in the unit cell (a=20.0165(3) Å, b=7.7594(10) Å, c=19.4809(3) Å, α=90 °, β=111.368(1) °, γ=90 ° and Z=8). Three-dimensional X-ray crystallographic structure of the compound has been determined and refined using SHELXS-97 and SHELXL-2014, respectively to a final R-value of 4.64%

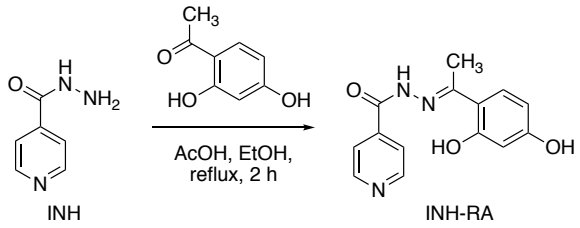
Keywords: Schiff's base; Isonicotinyl hydrazide; X-ray crystallography

1. Introduction

Isonicotinyl hydrazide (INH, Isoniazid) synthesis was first reported in 1912 but its antitubercular property was known only during 1942, it was then successfully introduced in to the market for the treatment tuberculosis in 1952 almost 40 years after its discovery.[1-3] Analogues of INH with antimycobacterial activity were then reported by many groups.[4] Schiff's bases of INH were also reported with their metal complexing ability and with wide range of biological activity.[5-10] We prepared a small library of schiff's bases of INH targeting DENV NS2B-NS3 protease (results to be published). Single crystal of schiff's base of INH with 2',4'-dihydroxy acetophenone (INH-RA) has been achieved and structure of the compound has been solved from its X-ray diffraction pattern.

2. Result and Discussion

Synthesis of the compound INH-RA has been achieved by the reaction outlined in the **Scheme 1**. Condensation of INH with 2',4'-dihydroxy acetophenone in the presence of catalytic amount of acetic acid provided the target compound. Crystallization was achieved through slow evaporation method. Proton NMR spectra of the compound displayed singlet for methyl protons in the region δ 2.42ppm. Amino and hydroxyl protons were also displayed singlet at δ 11.43, 9.94 & 13.39ppm. Aromatic protons of pyridyl and phenyl ring appeared as multiplets in the region between δ 6.28 and 8.79 ppm. ESI-MS displayed the molecular ion peak at 271.7 m/z.



Scheme 1. Synthesis of INH-RA

The compound crystallizes in the monoclinic C2/c space group with eight molecules in the unit cell (a=20.0165(3) Å, b=7.7594(10) Å, c=19.4809(3) Å, α=90 °, β=111.368(1) °, γ=90 ° and Z=8). The three dimensional molecular structure of this compound was determined by X-ray crystallography using SHELXS-97[11] and later refined by SHELXL-2014[12] to a final R-value 4.64%. Data pertaining to the single crystal and refinement are presented in Table 1. Bond length, bond angle and torsion angle (dihedral angle) date are summarized in Table 2-4, respectively.

In the unit cell contain 1:2 ratio of compound and water molecules are present. In the two water molecule, the one water molecule is half of the molecule in an asymmetric unit; the complete molecule is generated by crystallographic inversion symmetry with the inversion symmetric code of 1-x, y, 3/2-z. The pyridine ring is planar conformation with maximum deviation of atom C1 0.017(2) Å. The dihedral angle between the pyridine (N1-C1-C5) and the phenyl ring (C9-C14) is 29.29 (8) °. The propan acetohydrazide group is attached to the pyridine and the phenyl rings. The propan acetohyrazide group is *extended* conformation which can be seen from the torsion angle value [C3-C6-N2-N3] = 174.5 °, [C6-N2-

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N3-C7] = 177.1 ° and [N2-N3-C7-C9] = 177.5 °. The carbonyl group (O1) is oriented *syn-periplanar* to C10 [C8—C7—C9—C10 = -1.6(2) °] and *anti-periplanar* to C14 [C8—C7—C9—C14 = 177.1(1) °]. The methyl group (C8) is oriented *syn-periplanar* to C4 [O1—C6—C3—C4 = -24.6(2) °] and *anti-periplanar* to C2 [O1—C6—C3—C2 = 155.4(1) °]. The oxygen O2 and O3 atoms deviated from the phenyl ring (C9-C14) is -0.0043(13) Å and 0.0007(13) Å, respectively.

Table1. Crystal data and structure refinement

Parameters	Values
Empirical formula	C_{14} H_{16} N_3 $O_{4.50}$
Formula weight	298.3
Temperature	293(2) K
Wavelength	0.71073 Å
Crystal system, space group	Monoclinic, C 2/c
Unit cell dimensions	a = 20.0165(3) Å alpha = 90 °. b = 7.75940(10) Å beta = 111.3680(10) °. c = 19.4809(3) Å gamma = 90 °.
Volume	2817.71(7) A^3
Z, Calculated density	8, 1.406 Mg/m^3
Absorption coefficient	0.107 mm^{-1}
F(000)	1256
Crystal size	0.300 x 0.250 x 0.200 mm^3
Theta range for data collection	2.185 to 28.284 °.
Limiting indices	-26<=h<=21, -10<=k<=8, -24<=l<=25
Reflections collected / unique	13011 / 3496 [$R_{(int)}$ = 0.0304]
Completeness to theta = 25.242	100.00%
Refinement method	Full-matrix least-squares on F^2
Data / restraints / parameters	3496 / 0 / 210
Goodness-of-fit on F^2	1.036
Final R indices [I>2sigma(I)]	R1 = 0.0464, wR2 = 0.1160
R indices (all data)	R1 = 0.0792, wR2 = 0.1328
Largest diff. peak and hole	0.203 and -0.225 $e.A^{-3}$

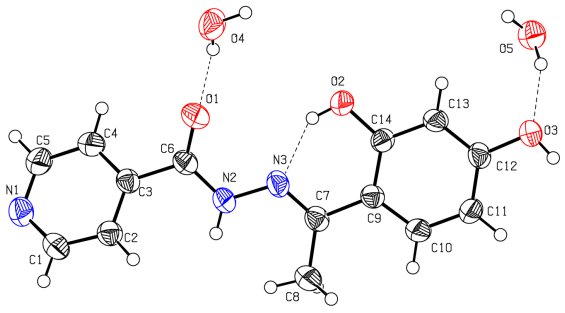

Figure 1. The ORTEP plot of the title compound with the atom numbering scheme. Displacement ellipsoids drown at 50 % probability level. The intramolecular interactions are shown in thin dashed line.

Intramolecular and intermolecular hydrogen bonds play a crucial role in stabilizing the structure and their packing, respectively (Table 5). Crystal structure is stabilized by O—H…N and O—H…O intramolecular hydrogen bonds as shown in Figure 1. In the crystal, the packing is stabilized by intermolecular N—H...O, O—H…O, O—H…N and C—H…O types of hydrogen bonds. In the crystal packing the water molecule atom O5 interacting with the N2—H2A…O5 type of intermolecular interaction. The O4 water molecule is behaving like donor and acceptor in the crystal packing of O3—H3…O4 and O4—H4A…N1 intermolecular interactions as shown in Figure 2. The intermolecular C4—H4...O1 hydrogen bond form a cyclic centrosymmetric $R^2_2(10)$ ring motif as shown in Figure 3. The C5—H5…O2 intermolecular interaction form a cyclic centrosymmetric $R^2_2(22)$ ring motif as shown in Figure 3. The C4—H4...O1 and C5—H5…O2 hydrogen bonds generating $R^2_2(12)$ ring motif viewed down *"b"* axis as shown in figure 3. The atom O2 is the bifurcated acceptor hydrogen bonds. In the crystal packing, the C1—H1…O2 intermolecular interaction forming a infinity chain (*C11*) running along (001) plane is shown in Figure 3.

Table 2. Selected Bond lengths [Å]

Bond	Bond Length (Å)
C1—N1	1.329(2)
C1—C2	1.382(2)
C2—C3	1.383(2)
C3—C4	1.375(2)
C3—C6	1.498(2)
C4—C5	1.377(2)
C5—N1	1.328(2)
C6—O1	1.2213(18)
C6—N2	1.347(2)
C7—N3	1.289(2)
C7—C9	1.470(2)
C7—C8	1.489(2)
C9—C10	1.396(2)
C9—C14	1.407(2)
C10—C11	1.375(2)
C11—C12	1.383(2)
C12—O3	1.3668(18)
C12—C13	1.379(2)
C13—C14	1.387(2)
C14—O2	1.3564(18)
N2—N3	1.3793(17)
O4—H4A	0.88(3)
O4—H4B	0.77(3)
O5—H5A	0.86(2)

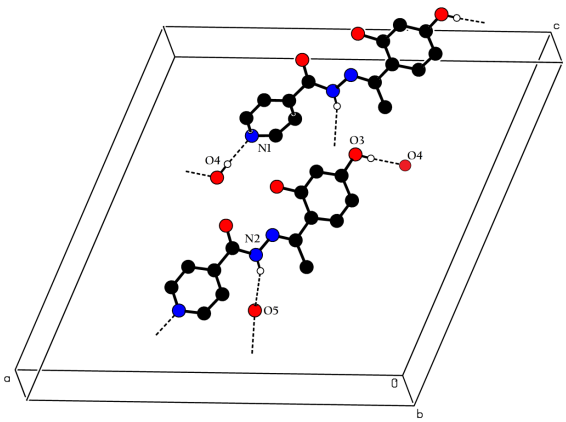

Figure 2. The N2—H2A…O5, O3—H3…O4 and O4—H4A…N1 intermolecular interactions viewed down *"b"* axis.

3. Conclusion

Three dimensional X-ray crystallographic structure of INH-RA has been solved successfully from the monoclinic crystals obtained on slow evaporation of compound from methanol. Hydroxyl functional group at 2nd position of the phenyl ring could able to establish both intra and intermolecular hydrogen bonding as methyl group of acetophenone lies opposite to it. While

intramolecular hydrogen bonding aids in stabilizing the structure, intermolecular hydrogen bonding was helping in crystal packing. Carbonyl oxygen of INH was found to establish second intermolecular hydrogen bonding that aids crystal packing. These information are vital for evaluating binding affinity of this compound with NS2B-NS3 protease of Dengue virus through computational modeling studies.

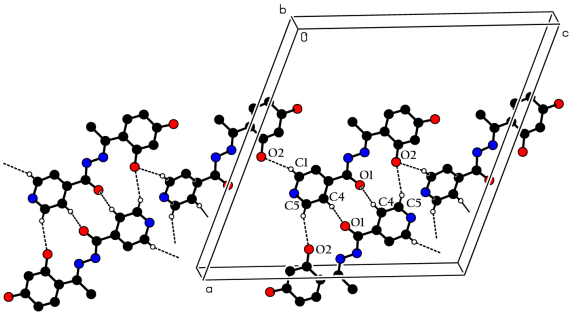

Figure 3. The C—H…O intermolecular hydrogen bonds generating $R^2_2(10)$, $R^2_2(22)$ and $R^2_2(12)$ ring motifs viewed down *"b"* axis.

Table 3. Selected Bond angles [°]

Bond	Bond Angle (°)
N1—C1—C2	123.66(16)
C1—C2—C3	118.56(15)
C4—C3—C2	118.11(14)
C4—C3—C6	118.65(14)
C2—C3—C6	123.24(14)
C3—C4—C5	119.08(16)
N1—C5—C4	123.58(17)
O1—C6—N2	123.11(15)
O1—C6—C3	121.58(15)
N2—C6—C3	115.30(14)
N3—C7—C9	115.58(14)
N3—C7—C8	124.02(14)
C9—C7—C8	120.39(14)
C10—C9—C14	116.59(14)
C10—C9—C7	121.15(14)
C14—C9—C7	122.24(14)
C11—C10—C9	122.98(15)
C10—C11—C12	118.94(15)
O3—C12—C13	117.56(14)
O3—C12—C11	122.13(14)
C13—C12—C11	120.31(14)
C12—C13—C14	120.30(14)
O2—C14—C13	116.48(13)
O2—C14—C9	122.64(13)
C13—C14—C9	120.87(14)
C5—N1—C1	116.93(15)
C6—N2—N3	117.44(13)
C7—N3—N2	120.14(13)
H4A—O4—H4B	112(2)

4. Experimental

3.1. Synthesis of N'-[(1E)-1-(2,4-dihydroxyphenyl)ethylidene]pyridine-4-carbohydrazide (INH-RA): To a solution of isonicotynylhydrazide (0.5 g, 3.65 mmol) in 40 mL of ethanol, added 1-(2,4-dihydroxyphenyl)-ethanone (0.55g; 3.65 mmol). To this mixture added 2-3 drops of glacial acetic acid and refluxed for 2 hours. After the completion of the reaction, monitored through TLC, the organic layer was removed in vacuo, washed with water, filtered and dried to provide the target compound INH-RA. ^{1}HNMR (400 MHz) δ ppm: 2.42 (s, 3H, -CH_3); 6.28-7.47 (m, 3H, Ar-H); 7.83 (d, 2H, J=6.0 Hz, Pyr-H); 8.79 (d, 2H, J=6.0 Hz, Pyr-H); 9.94 (s, 1H, -OH); 11.43 (s, 1H, -NH); 13.39 (s, 1H, -OH). ESI-MS (m/z): 271.7 $[M]^+$

Table 4. Torsion angle

Bond	Torsion Angle (°)
N1—C1—C2—C3	2.4(3)
C1—C2—C3—C4	0.5(2)
C1—C2—C3—C6	-179.52(15)
C2—C3—C4—C5	-2.3(3)
C6—C3—C4—C5	177.65(16)
C3—C4—C5—N1	1.7(3)
C4—C3—C6—O1	34.6(2)
C2—C3—C6—O1	-145.39(17)
C4—C3—C6—N2	-144.10(16)
C2—C3—C6—N2	35.9(2)
N3—C7—C9—C10	178.48(14)
C8—C7—C9—C10	-1.6(2)
N3—C7—C9—C14	-2.8(2)
C8—C7—C9—C14	177.14(16)
C14—C9—C10—C11	0.1(2)
C7—C9—C10—C11	178.89(15)
C9—C10—C11—C12	-0.2(3)
C10—C11—C12—O3	179.88(15)
C10—C11—C12—C13	0.5(2)
O3—C12—C13—C14	179.79(13)
C11—C12—C13—C14	-0.8(2)
C12—C13—C14—O2	-179.97(14)
C12—C13—C14—C9	0.8(2)
C10—C9—C14—O2	-179.63(14)
C7—C9—C14—O2	1.6(2)
C10—C9—C14—C13	-0.4(2)
C7—C9—C14—C13	-179.16(13)
C4—C5—N1—C1	1.0(3)
C2—C1—N1—C5	-3.0(3)
O1—C6—N2—N3	-4.2(2)
C3—C6—N2—N3	174.55(12)
C9—C7—N3—N2	177.50(13)
C8—C7—N3—N2	-2.5(2)
C6—N2—N3—C7	177.10(14)

Table 5. Hydrogen Bonds

D—H…A	D—H (Å)	H…A (Å)	D…A (Å)	D—H…A [°]
N2-H2A…O5 (i)	0.86	2.41	3.1581(13)	146
O2-H2B…N3	0.82	1.82	2.5357(17)	145
O3-H3…O4 (ii)	0.82	1.79	2.614(2)	176
O4-H4A…N1 (iii)	0.89(3)	1.92(3)	2.803(2)	173(2)
O4-H4B…O1	0.77(3)	2.09(3)	2.844(2)	167(3)
O5-H5A…O3	0.86(2)	2.03(2)	2.8520(17)	158(2)
C1-H1…O2 (iv)	0.93	2.52	3.414(2)	160
C4-H4…O1 (v)	0.93	2.52	3.383(2)	155
C5-H5…O2 (v)	0.93	2.58	3.449(2)	156

Symmetry codes: i) 1-x, 1-y, 1-z; ii) -1/2+x, -1/2+y, z; iii) x, 2-y, ½+z; iv) x, 2-y, -1/2+z; v) 3/2-x, 3/2-y, 1-z.

3.2. Crystallization: Compound INH-RA was purified by means of recrystallization from hot methanol. Recrystallization solvent methanol has been used for obtaining single crystal of INH-RA through slow evaporation method.

3.3. X-ray analysis: X-ray diffraction intensity data were collected at room temperature (293K) on a Bruker AXS SMART APEXII[13] single crystal X-ray diffractometer equipped with graphite monochromatic Mo$K\alpha$ (λ=0.71073 Å) radiation and CCD detector. A crystal of

dimensions 0.30 X 0.25 X 0.20 mm^3 was mounted on a glass fiber using cyanoacrylate adhesive. The unit cell parameters were determined from 36 frames measured (0.5° phi-scan) from three different crystallographic zones using the method of difference vectors. The intensity data were collected with an average four-fold redundancy per reflection and optimum resolution (0.75 Å). The intensity data collection, frames integration, Lorentz and polarization corrections and decay correction were carried out using *SAINT-NT* (version 7.06a) software. An empirical absorption correction (multi-scan) was performed using the *SADABS* program. The crystal structure was solved by direct methods using *SHELXS-97* and refined by full-matrix least-squares using *SHELXL-2014*. Molecular geometry was calculated using PARST[14-15]. All non-hydrogen atoms were refined using anisotropic thermal parameters. The hydrogen atoms were included in the structure factor calculation at idealized positions by using a riding model, but not refined. Images were created with the ORTEP-PLATON program[16-17].

Refinement: The hydrogen atoms were placed in calculated positions with C—H = 0.93 Å to 0.96 Å, N—H = 0.86 Å and O—H = 0.77 Å to 0.88 Å refined in the riding model with fixed isotropic displacement parameters: U_{iso}(H) = 1.5*U*eq(C) for methyl groups and U_{iso}(H) = 1.2*U*eq(C) for C aromatic. The methyl groups were allowed to rotate but not to tip.

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Supplementary Information

CCDC 1570752 contains the supplementary crystallographic data of Compound INH-RA. This data can be obtained free of charge from the Cambridge Crystallographic Centre via www.ccdc.cam.ac.uk/data_request/cif.

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