Electronic supplementary materials *Mendeleev Commun.,* 2024, **34**, 850–853

Insight into design of 3-hydroxy-4-pyridinone functionalized with isoniazid fragment: structural characterization and antimycobacterial evaluation

Marina A. Katkova, Galina S. Zabrodina, Roman V. Rumyantcev, Grigory Yu. Zhigulin, Ivan V. Skabitsky, Irina G. Fomina, Olga B. Bekker, Sergey Yu. Ketkov and Igor L. Eremenko

1. Experimental

General Procedures

All chemicals were reagent-grade and were used as received from Sigma Aldrich without any additional purification. The elemental analyses were carried out on a EuroVector CHNS-O Elemental Analyzer Euro EA 3000. ^{1}H NMR spectrum was recorded on Bruker Avance 300 spectrometer (300 MHz for ^{1}H). Chemical shifts are given relative to $SiMe_4$ and are referenced to residual signal of DMSO-d_6 (δ_H 2.50). The electronic spectra of the complexes were recorded on a Perkin–Elmer Lambda 25 UV/Vis spectrometer (range: 220–1100 nm) at ambient temperature. IR spectra (IRS) were recorded on a Perkin Elmer Spectrum 65 spectrophotometer equipped with a Quest ATR Accessory (Specac) by the attenuated total reflectance (ATR) in the range of 400–4000 cm^{-1}. Microprobe analyses were carried out using a Carlo Erba EA 1108 Series CHNS Elemental Analyzer.

Biological activity of compounds was determined in '*M. smegmatis* mc2 155' test systems by paper disk-diffusion method. A culture of *M. smegmatis* was grown as a lawn on an agar medium around paper disks with a diameter of 6 mm, soaked in solutions of the test compound in DMSO, having different concentrations. Bacteria grown on Tryptone soya agar M-290 (Himedia) were cultivated for a night in Lemco-TW liquid medium (Lab Lemco' Powder 5 g/L (Oxoid), Peptone special 5 g/L (Oxoid), NaCl 5 g/L, Tween-80) at 37 °C until logarithmic mean growth phase at optical density OD600 = 1.5. Bacteria was mixed with melt agar medium M-290 in ratio 1 : 9 : 10 (culture : Lemco-TW : M-290) and poured as the top layer onto Petri dishes with lower agar M-290. After the top layer had hardened, paper disks soaked in the test substances were placed on the surface. The culture was incubated for 24 h at 37°C. The diameter of the growth inhibition zone (halo) of a *M. smegmatis* culture grown in a lawn on an agar medium around paper disks containing the test compound in different concentrations was quantified. The minimum inhibitory concentration (MIC) was considered to be the concentration of the compound at which the growth inhibition zone is minimal.

Synthesis of N-(3-hydroxy-2-methyl-4-oxo-4H-pyridin-1-yl)isonicotinamide (1)

3-Hydroxy-2-methyl-4*H*-pyran-4-one (20.18 g, 0.16 mol) and pyridine-4-carbohydrazide (21.94 g, 0.16 mol) were suspended in 200 ml of hot water, and the reaction mixture was refluxed under argon for 48 h. The mixture was allowed to cool and the beige solid was separated by filtration, washed repeatedly with hot water and hot methanol and recrystallized from water. Slow evaporation of the solvent yielded 13.6 g (35 %) of beige crystals. Anal. Calcd. for $C_{12}H_{11}N_3O_3 \cdot 2H_2O$ (**1**): C, 51.24; H, 5.38; N, 14.94 %. Found: C, 51.20; H, 5.43; N, 14.91 %. ^{1}H NMR (300 MHz, DMSO-d_6): δ = 2.14 (s, 3H, CH_3), 6.21 (d, J = 7.6 Hz, 1H, CH), 7.75 (d, J = 7.6 Hz, 1H, CH), 7.85 (d, J = 5.4 Hz, 2H, C_5NH_4), 8.84 (d, J = 5.4 Hz, 2H, C_5NH_4), 12.40 (s, 1H, NH). UV-vis λ_{max}, nm (ε, dm^3 mol^{-1} cm^{-1}) in MeOH at 298 K: 264 (12000). IR spectra (ν,

cm^{-1}): 404.28 w, 421.79 w, 494.61 s, 521.55 s, 614.3 s, 638.89 s, 695.99 m, 753.64 m, 795.54 s, 828.39 s, 898.07 s, 916.79 s, 999. 33 m, 1026.57 s, 1063.11 m, 1108.15 m, 1189.0 s, 1217.44 s, 1272.15 s, 1312.81 s, 1377.64 m, 1415.75 s, 1483.92 s, 1548.55 s, 1572.50 m, 1625.17 m, 1415.75 s, 2927.41 w.

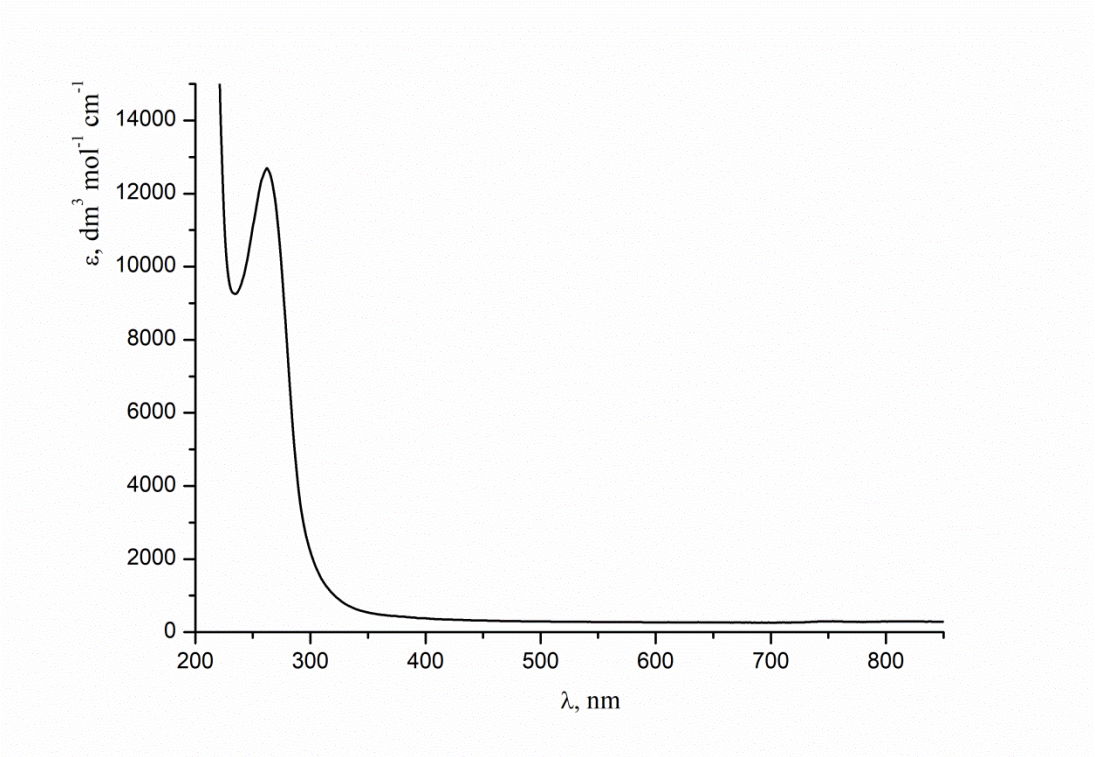

Figure S1. UV/Vis spectrum of *N-(3-hydroxy-2-methyl-4-oxo-4H-pyridin-1-yl)-isonicotinamide (1)*

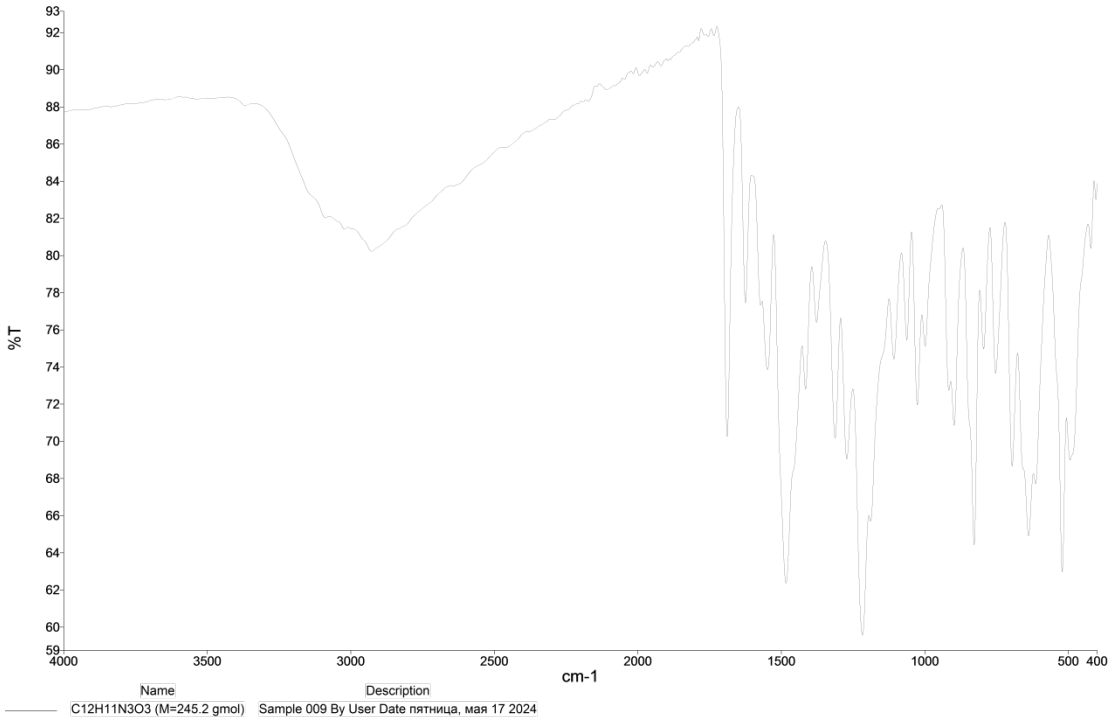

Figure S2. IR spectrum of *N-(3-hydroxy-2-methyl-4-oxo-4H-pyridin-1-yl)isonicotinamide (1)*

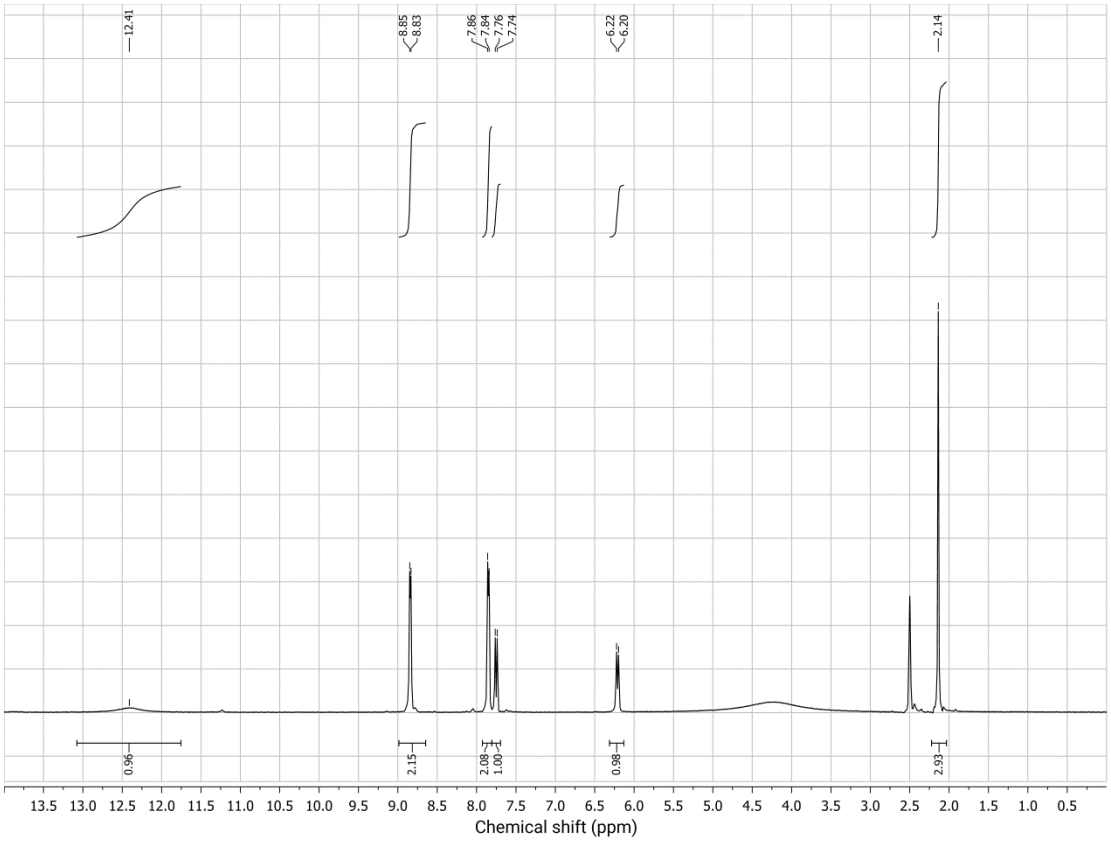

Figure S3. ^{1}H NMR spectrum of *N-(3-hydroxy-2-methyl-4-oxo-4H-pyridin-1-yl)-isonicotinamide (1)* in DMSO-d_6

Table S1 The selected bond lengths (Å) and angles (deg) in the compound **1**.

Bond	**1**, [Å]	Bond	**1**, [Å]	Angle	**1**, [deg]
O1-C1	1.2744(11)	N2-C7	1.3605(12)	C4-N1-C3	122.74(8)
O2-C2	1.3625(11)	C1-C2	1.4336(12)	C4-N1-N2	119.33(8)
O3-C7	1.2189(12)	C2-C3	1.3686(12)	C3-N1-N2	117.91(8)
N1-N2	1.3927(10)	C4-C5	1.3617(13)	C7-N2-N1	119.17(8)
N1-C3	1.3782(12)	C1-C5	1.4216(13)	C7-N2-H2B	124.8(10)
N1-C4	1.3545(12)	O2-H2A	0.887(18)	N1-N2-H2B	115.7(10)
N3-C10	1.3394(15)	N2-H2B	0.889(16)	C11-N3-C10	117.73(9)
N3-C11	1.3371(14)	C3-C6	1.4909(13)	C2-O2-H2A	112.1(12)

Table S2 Geometric characteristics of O–H**…**O and N–H**…**O interactions in the crystal **1**.

D–H…*A*	*D*–H, Å	H…*A*, Å	*D*…*A*, Å	∠*D*H*A*, град
O2–H2A...O1$^{\#1}$	0.887(18)	1.830(18)	2.6330(10)	149.6(16)
N2–H2B...O2S$^{\#2}$	0.889(16)	1.823(17)	2.7060(11)	171.8(14)
O1S–H1SA...O2	0.87(2)	2.04(2)	2.8961(11)	164.8(17)
O1S–H1SB...N3$^{\#3}$	0.879(19)	1.92(2)	2.7932(11)	172.6(17)
O2S–H2SA...O1	0.879(18)	1.836(18)	2.7010(10)	167.8(17)
O2S–H2SB...O1S$^{\#4}$	0.894(19)	1.861(19)	2.7431(10)	168.5(17)

Symmetry transformations used to generate equivalent atoms: $^{\#1}$ –x+1, –y+2, –z+1; $^{\#2}$ –x+1, –y+1, –z+1; $^{\#3}$ x, y+1, z+1; $^{\#4}$ x–1, y, z.

Table S3 Interatomic distances (*d*) and values of the electron density ρ(r) at the bond critical points (3,–1) for DFT optimized structures of INH and **1**.

Bond	INH		**1**	
	d, Å	ρ(**r**), a.u.	*d*, Å	ρ(**r**), a.u.
O=C(N)	1.216	0.412	1.207	0.420
N–N	1.383	0.340	1.364	0.356
(O)C–N(N)	1.355	0.332	1.370	0.318