A New Complex of Diperoxovanadate with 2-(2′-pyridyl)-imidazole: Synthesis, Structure, Interaction and Bioactivity

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Introduction

Vanadate compounds were reported to be a new kind of powerful insulin mimic and anticancer agents based on their biological response both *in vitro* and *in vivo*. A novel diperoxovanadate complex $NH_4[OV(O_2)_2\{2\text{-}(2'\text{-pyridyl})\text{-imidazole}\}]\cdot 4H_2O$ was synthesized in aqueous solution under physiological conditions. Its solution structure was characterized by multinuclear (^{1}H, ^{13}C, ^{14}N, and ^{51}V) as well as multi-dimensional (DOSY and C-H COSY) NMR techniques in the interaction system of NH_4VO_3/H_2O_2/2-(2′-pyridyl)-imidazole at room temperature. It affected the proliferation of tumor cell and normal one in a different manner. In addition, it inhibited the specific activity of ALP (alkaline phosphatase) in gastric cancer cell MGC-803.

Methods

$NH_4[OV(O_2)_2\{2\text{-}(2'\text{-Py})\text{-Imi}\}]\cdot 4H_2O$ was synthesized according to literature [1]. Anal. Calcd. for $NH_4[OV(O_{2)2}(C_8H_7N_3)]\cdot 4H_2O$ ($C_8H_{19}N_4O_9V$): C, 26.24%; H, 5.23%; N, 15.30%. Found: C, 26.11%; H, 5.05%; N, 15.41%. IR (KBr): υ = 3498 vs (>N–H), 3195 vs, 2916 vs, 2757 vs (H_3N^+–H), 1654 m, 1477 s, 1453 m, 1401 m, 1291 w, 1159 m, 974 m, 940 s (V=O), 923 m, 882 m $(O–O)_{peroxo}$, 863 s $(O–O)_{peroxo}$, 790 m, 750 m, 707 m, 627 m $(V–O_{peroxo})$, 588 s $(V–O_{peroxo})$ cm^{-1}. Raman: υ= 974 s (V=O), 884 s $(O–O)_{peroxo}$, 632 m $(V–O_{peroxo})$, 588 m $(V–O_{peroxo})$, 494 vs $(V–O_{peroxo})$ cm^{-1}. ^{1}H NMR (D_2O): 7.26 (t, J = 5.9 Hz, 1H), 7.47 (s, 1H), 7.67 (d, J = 7.7 Hz, 1H), 7.75 (s, 1H), 7.82 (t, J = 7.7 Hz, 1H), 8.05 (s, 1H) ppm. ^{13}C NMR (D_2O): 149.0, 147.4, 145.1, 141.1,132.9, 127.0, 123.9, 121.2 ppm.

All NMR spectra were recorded on Varian Unity plus 500 spectrometer operating at 500.4 MHz for ^{1}H, 125.7 MHz for ^{13}C, 36.12 MHz for ^{14}N, and 131.4 MHz for ^{51}V NMR. The solvent was D_2O. DOSY was recorded using a z-gradient probe, which delivered a maximum gradient strength of 30 G/cm. The DOSY gradient compensated stimulated echo spin lock (DGCSTESL) sequence [2] was used for acquiring DOSY spectra. The crystal structure of the complex was determined at 173 K by single-crystal X-ray diffraction method. Cell proliferation assay was carried out as described in literature [3]. ALP activity assay was applied according to Daniel A. Barrio [4] with some modifications.

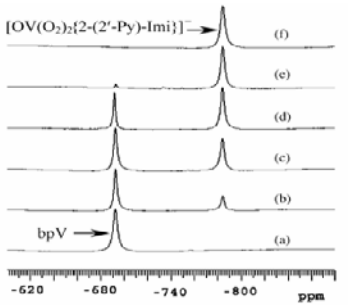

Fig. 1. ^{51}V spectra of the NH_4VO_3/H_2O_2/2-(2′-Py)-Imi aqueous solution with 0.2 mol/L the total concentration of vanadate species.

Results and Discussion

When 0.25, 0.5, 0.75, 1.0, or 2.0 equivalent of 2-(2′-Py)-Imi was added to the NH_4VO_3/H_2O_2 (1:5 molar ratio) aqueous solution (Fig. 1(a), where bpV refers to the species $[OV(O_2)_2(H_2O)]/[OV(O_2)_2(D_2O)]$), a new single peak appeared at about -784 ppm, as shown in Fig. 1(b)~(f), which was assigned to a new species $[OV(O_2)_2\{2\text{-}(2'\text{-Py})\text{-Imi}\}]^-$. Its chemical shift is at the highest field among the known mononuclear diperoxovanadate(V) compounds. The intensity of the new peak increased with the increasing of the quantity of ligand before reaching a maximum.

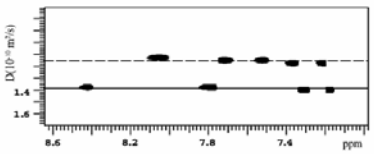

Fig. 2. 2D ^{1}H DOSY spectrum of the NH_4VO_3/H_2O_2/2-(2′-Py)-Imi with a molar ratio of 1:5:2 and pH 7.6 in aqueous solution.

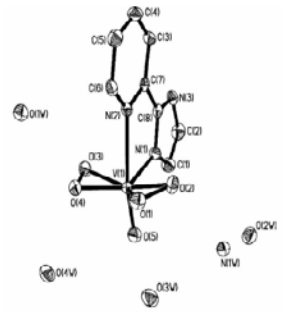

Fig. 3. Structure of $NH_4[OV(O_2)_2\{2\text{-}(2'\text{-Py})\text{-Imi}\}]\cdot 4H_2O$.

The ^{1}H DOSY spectrum of NH_4VO_3/H_2O_2/2-(2′-Py)-Imi (1:5:2, 0.2 mol/L vanadate concentration) in solution is shown in Fig. 2. The component with slower diffusion rate marked by broken line was assigned to the newly-formed species, $[OV(O_2)_2\{2\text{-}(2'\text{-Py})\text{-Imi}\}]^-$. Another component marked by solid lines was assigned to the free ligand.

The structure unit of the crystal is shown in Fig. 3. The species, $[OV(O_2)_2\{2\text{-}(2'\text{-Py})\text{-Imi}\}]^-$, has a distorted pentagonal bipyramidal geometry, as often observed in transition metal peroxo complexes.

Inhibition of $NH_4[OV(O_2)_2\{2\text{-}(2'\text{-Py})\text{-Imi}\}]$ on cell proliferation was determined by MTT colorimetric assay. The IC_{50} measurements of $NH_4[OV(O_2)_2\{2\text{-}(2'\text{-Py})\text{-Imi}\}]$ were 30.9μM in L342, 7.35μM in MGc-803, 6.2μM in 95D, and 71.5μM in HEK293. It shows a higher toxicity against some of the cancer cell lines tested, compared with that in the control cell lines HEK293. The effect of $NH_4[OV(O_2)_2\{2\text{-}(2'\text{-Py})\text{-Imi}\}]$ on ALP activity was measured by p-NPP colorimetric assay. As shown in Fig. 4, the new complex in the range of 0.1-1000μM had an inhibitory effect of more than 60% on the ALP activity of cell extracted from gastric cancer cell line MGC-803.

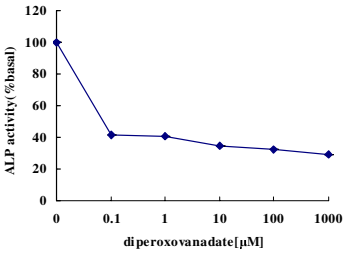

Fig. 4. Effects of new complex on ALP activities extracted from gastric cancer cell MGC-803. Cell extract added to the glycine buffer with 5mM p-NPP alone (basal) or with different concentrations of the new complex.

Acknowledgments

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