Electronic Supplementary Information for Dalton Transactions

Electronic Supplementary Information

Vanadium complexes having $[V^{IV}O]^{2+}$ and $[V^{V}O_2]^{+}$ cores with binucleating dibasic tetradentate ligands: Synthesis, characterization, catalytic and antiamoebic activities

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Experimental

In vitro testing against E. histolytica

The ligands and their dioxidovavandium(V) complexes (**2**, **3**, **5** and **6**) were screened in vitro for antiamoebic activity against *HM1:1MSS* strain of *E. histolytica* by using a microplate method.[35] *E. histolytica* trophozoites were cultured in TYIS-33 growth medium in wells of 96 microtiter plate (Costar).[36] DMSO (40 μL) was added to all the samples (1 mg) followed by enough culture medium to obtain concentration of 1 mg/mL. The maximum concentration of DMSO in the tests did not exceed 0.1%; at this level no inhibition of amoebal growth occurred.[37,38] ^{1}H-NMR spectra of all complexes in DMSO-d_6 were recorded to test whether the solvent induced any hydrolysis, but all were found to be stable at room temperature for several days. Samples were dissolved or suspended by mild sonication for few minutes and then further dilution with medium to concentration of 0.1 mg/mL. Two fold serial dilutions were made in the wells of the 96-well microtiter plate in 170μL of the medium. Each test included metronidazole as the standard amoebicidal drug, control wells (culture medium plus amoebae) were prepared from a confluent culture by pouring off the medium, adding 2 mL of medium and chilling the culture on ice to detach the organisms from the side of the flask. The number of the amoeba per mL was estimated with a heamocytometer and trypan blue exclusion was used to confirm viability. The cell suspension used was diluted to 10^5 organisms/ mL by adding fresh medium and 170μL of this suspension was added to the test and control well in the plate so that the wells were completely filled (total volume, 340 μL). An inoculum of 1.7×10^4 organisms/ well was chosen so that confluent, but not excessive growth took place in control wells. The plate was sealed with expanded polystyrene (0.5 thick). Secured with tape, placed in

a modular incubating chamber (flow laboratories, High Wycombe, UK), and gassed for 10 min with nitrogen before incubation at 37 ºC for 72 h.

Assessment of antiamoebic activity

After incubation, the growth of amoebae in the plate was checked with a low power microscope. The culture medium was removed by inverting the plate and shaking gently. The plate was then immediately washed once in NaCl solution (0.9 %) at 37 ºC. This procedure was completed quickly, and the plate was not allowed to cool to prevent the detachment of amoebae. The plate was allowed to dry at room temperature, and the amoebae was fixed with methanol, when dry, stained with (0.5%) aqueous eosin for 15 min. Stained plate was washed once with tap water and then twice with distilled water and allowed to dry. A 200 µL portion of 0.1 N sodium hydroxide solutions was added in each well to dissolve the protein and release the dye. The optical density of the resulting solution in each well was determined at 490 nm with a micro plate reader. The % inhibition of amoebal growth was calculated from the optical densities of the control and test wells and plotted against the logarithm of the dose of the drug tested. Linear regression analysis was used to determine the best-fitted straight line from which the IC_{50} value was found. (Note: For references see original text)

Table S1 Selected bond lengths (Å) and angles (º) for $[Cs]_2[CH_2\{V^VO_2(sal\text{-}bhz)\}_2]$ **3**

Cs(1)-O(2)#1	2.944(4)	V(1)-O(7)	1.622(5)
Cs(1)-O(6)#2	3.081(5)	V(1)-O(6)	1.639(5)
Cs(1)-O(2W)#3	3.118(5)	V(1)-O(5)	1.895(5)
Cs(1)-O(1)	3.131(5)	V(1)-O(8)	2.001(5)
Cs(1)-O(3)#3	3.164(5)	V(1)-N(3)	2.145(6)
Cs(1)-O(3)	3.170(5)	Cs(1)-V(2)#3	3.8426(12)
Cs(1)-O(2)#3	3.177(5)	Cs(1)-V(2)	3.8457(12)
Cs(1)-O(2W)#1	3.242(5)	V(1)-Cs(2)#4	3.7818(12)
Cs(1)-C(23)	3.703(6)	V(1)-Cs(2)#5	4.0020(12)
Cs(1)-C(24)	3.901(6)	V(2)-O(3)	1.616(5)
Cs(2)-O(2)#1	2.955(5)	V(2)-O(2)	1.644(5)
Cs(2)-O(5)#2	3.070(5)	V(2)-O(4)	1.907(5)
Cs(2)-O(8)#7	3.073(5)	V(2)-N(2)	2.144(6)
Cs(2)-O(7)#7	3.155(6)	V(2)-Cs(1)#9	3.8426(12)
Cs(2)-O(6)#2	3.192(5)	V(2)-Cs(2)#6	4.1059(12)
Cs(2)-O(2W)#8	3.261(5)	V(2)-Cs(1)#6	4.2578(11)
Cs(2)-C(28)	3.384(8)	Cs(2)-V(1)#7	3.7818(12)
Cs(2)-O(1)#1	3.430(5)	O(1)-V(2)	1.973(5)
Cs(2)-C(27)	3.519(8)	O(1)-C(23)	1.321(8)
Cs(2)-C(14)#2	3.842(6)	N(1)-C(23)	1.313(9)
Cs(2)-C(13)#2	3.851(7)	N(1)-N(2)	1.386(8)

O(2)-Cs(1)#6	2.944(4)	N(2)-C(22)	1.298(10)
O(2)-Cs(2)#6	2.955(5)	N(3)-C(8)	1.295(10)
O(2)-Cs(1)#9	3.177(5)	N(3)-N(4)	1.404(8)
O(2W)-Cs(1)#9	3.118(5)	N(4)-C(7)	1.289(10)
O(2W)-Cs(1)#6	3.242(5)	O(5)-C(14)	1.323(8)
O(2W)-Cs(2)#10	3.261(5)	C(7)-O(8)	1.321(8)
O(3)-Cs(1)#9	3.164(5)		
O(6)-Cs(1)#5	3.081(5)	O(2)#1-Cs(1)-C(23)	86.94(15)
O(6)-Cs(2)#5	3.192(5)	O(6)#2-Cs(1)-C(23)	91.48(14)
O(7)-Cs(2)#4	3.155(6)	O(2W)#3-Cs(1)-C(23)	109.59(13)
O(8)-Cs(2)#4	3.073(5)	O(1)-Cs(1)-C(23)	20.13(13)
C(13)-Cs(2)#5	3.851(7)	O(3)#3-Cs(1)-C(23)	166.13(14)
C(14)-Cs(2)#5	3.842(6)	O(3)-Cs(1)-C(23)	60.04(14)
		O(2)#3-Cs(1)-C(23)	144.03(14)
O(2)#1-Cs(1)-O(6)#2	89.29(14)	O(2W)#1-Cs(1)-C(23)	99.35(13)
O(2)#1-Cs(1)-O(2W)#3	145.29(13)	O(2)#1-Cs(1)-V(2)#3	105.87(9)
O(6)#2-Cs(1)-O(2W)#3	60.91(13)	O(6)#2-Cs(1)-V(2)#3	92.45(10)
O(2)#1-Cs(1)-O(1)	106.56(12)	O(2W)#3-Cs(1)-V(2)#3	61.76(8)
O(6)#2-Cs(1)-O(1)	87.29(13)	O(1)-Cs(1)-V(2)#3	147.56(8)
O(2W)#3-Cs(1)-O(1)	90.41(12)	O(3)#3-Cs(1)-V(2)#3	24.28(9)
O(2)#1-Cs(1)-O(3)#3	88.22(13)	O(3)-Cs(1)-V(2)#3	108.68(8)
O(6)#2-Cs(1)-O(3)#3	75.46(13)	O(2)#3-Cs(1)-V(2)#3	24.84(8)
O(2W)#3-Cs(1)-O(3)#3	68.37(12)	O(2W)#1-Cs(1)-V(2)#3	82.69(8)
O(1)-Cs(1)-O(3)#3	157.26(12)	C(23)-Cs(1)-V(2)#3	166.64(11)
O(2)#1-Cs(1)-O(3)	121.61(13)	O(7)-V(1)-O(6)	108.2(3)
O(6)#2-Cs(1)-O(3)	133.50(13)	O(7)-V(1)-O(5)	101.4(3)
O(2W)#3-Cs(1)-O(3)	92.86(13)	O(6)-V(1)-O(5)	97.0(2)
O(1)-Cs(1)-O(3)	52.90(12)	O(7)-V(1)-O(8)	98.9(2)
O(3)#3-Cs(1)-O(3)	132.92(9)	O(6)-V(1)-O(8)	92.5(2)
O(2)#1-Cs(1)-O(2)#3	117.65(13)	O(5)-V(1)-O(8)	153.6(2)
O(6)#2-Cs(1)-O(2)#3	113.19(13)	O(7)-V(1)-N(3)	112.8(2)
O(2W)#3-Cs(1)-O(2)#3	65.75(12)	O(6)-V(1)-N(3)	138.0(2)
O(1)-Cs(1)-O(2)#3	130.51(12)	O(5)-V(1)-N(3)	83.4(2)
O(3)#3-Cs(1)-O(2)#3	48.79(12)	O(8)-V(1)-N(3)	73.2(2)

Symmetry transformations used to generate equivalent atoms:

#1 x+1,y,z #2 -x+3/2,-y+1,z-1/2 #3 x+1/2,-y+1/2,-z

#4 -x+5/2,-y+1,z+1/2 #5 -x+3/2,-y+1,z+1/2 #6 x-1,y,z

#7 -x+5/2,-y+1,z-1/2 #8 x+3/2,-y+1/2,-z #9 x-1/2,-y+1/2,-z

#10 x-3/2,-y+1/2,-z

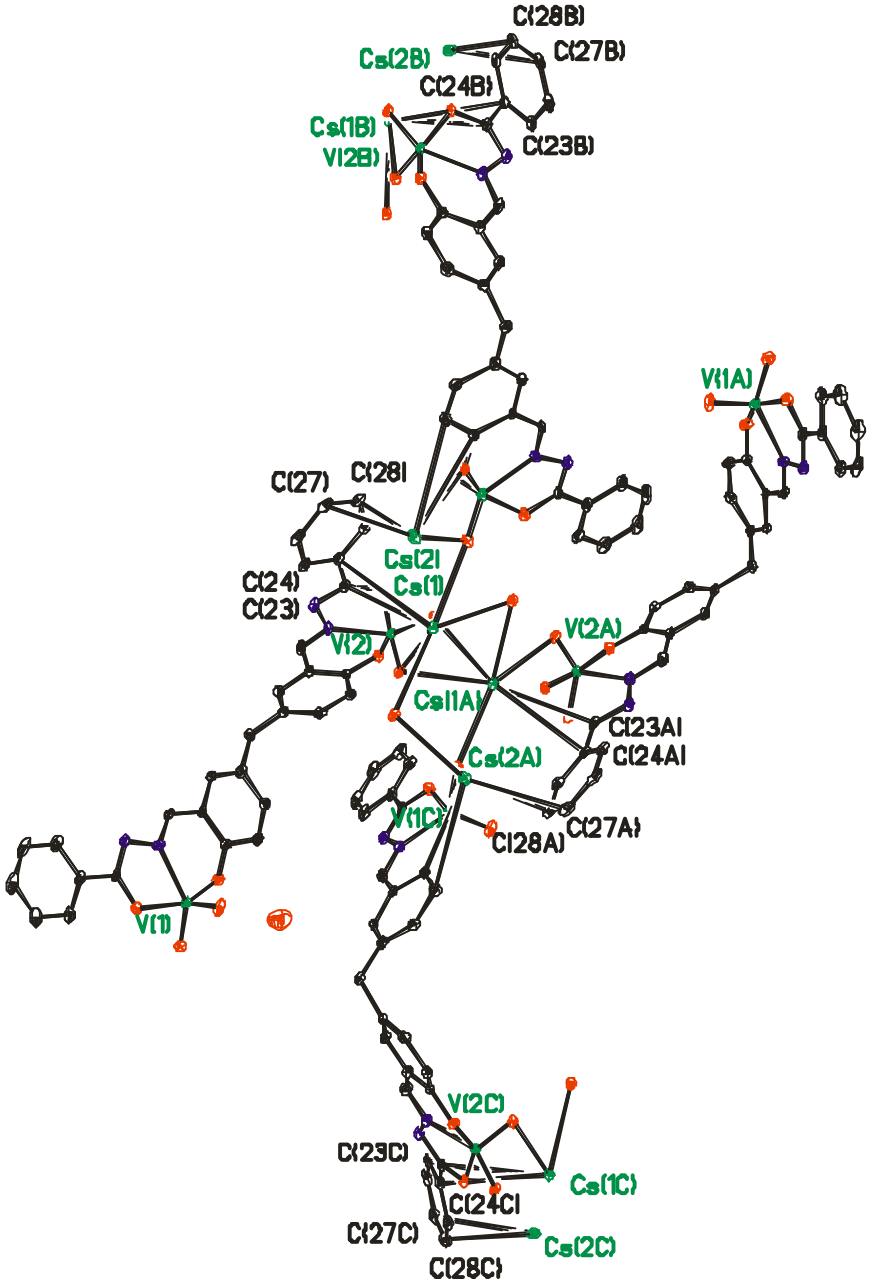


Fig. S1 The four dinuclear molecules present in the unit cell of compound **3**.

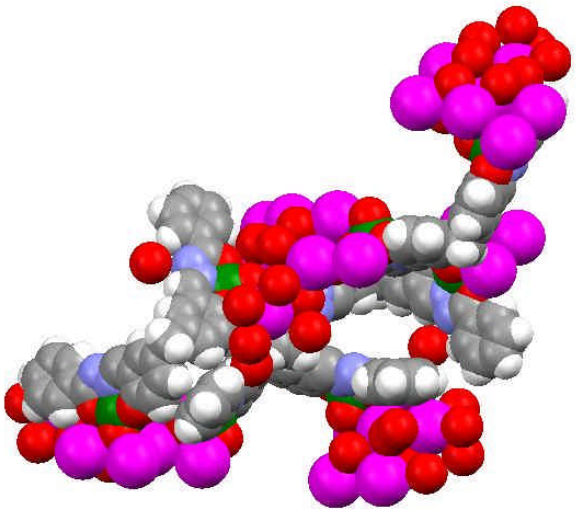
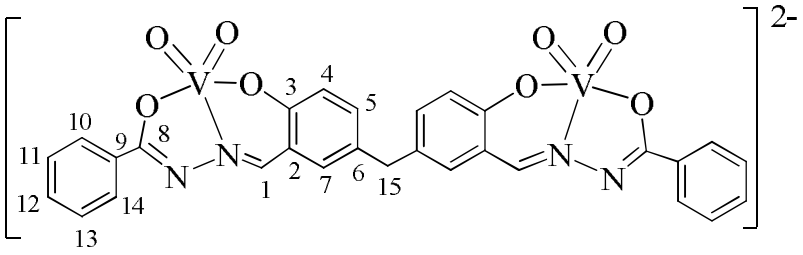


Fig. S2 Space filling representation of the crystal packing of compound **3**, showing cation π-interactions. In red: oxygen, pink: caesium, gray: carbon, green: vanadium, dark blue: nitrogen and white: hydrogen atoms.

^{13}C NMR studies

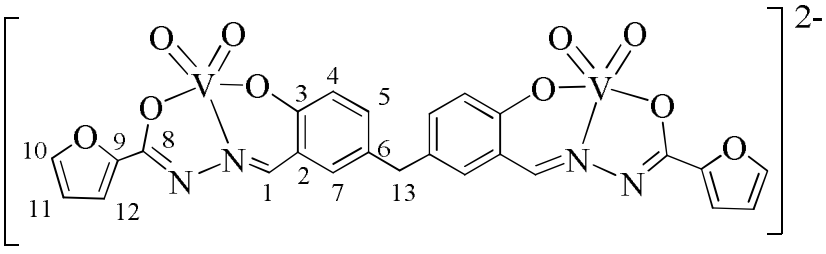
The ^{13}C spectra of complexes **2** and **3** contain 13 signals corresponding to the 27 carbon atoms of the molecules owing to their symmetry. The assignment of the peaks (Table S2) was made with the help of the Chem Draw® software and all of them match within ± 2 % accuracy with the predicted values. The peaks of atoms C14 overlap those of DMSO so they cannot be properly assigned. For compounds **5** and **6**, 10 and 9 distinct signals, respectively, were recorded (see Table S3)

Table S2 ^{13}C NMR chemical shifts observed; for the atom labelling see scheme below



Compound	C8	C1	C3	C10/11	C12/13	C14	C7/9	C5	C2	C4
$[K]_2[CH_2\{V^VO_2(sal\text{-}bhz)\}_2]$ **2**	163.5	148	147.9	145	134	132	130	120	113.7	112
$[Cs]_2[CH_2\{V^VO_2(sal\text{-}bhz)\}_2]$ **3**	163	147	142	134.9	132.3	130.4	121.9	120	119.7	

Table S3 ^{13}C NMR chemical shifts observed; for the atom labelling see scheme below



$[CH_2\{V^VO_2(sal\text{-}fah)\}_2]^{2-}$

Compound	C8	C1	C3	C9	C7/6/4/5	C10/11/12
$[K]_2[CH_2\{V^VO_2(sal\text{-}fah)\}_2]$ **5**	163.5	156	147.9	145	134 132 130	120 113.7 112
$[Cs]_2[CH_2\{V^VO_2(sal\text{-}fah)\}_2]$ **6**	163.5	156	147	145	134 132	119 113 112

UV-Vis studies (additional information)

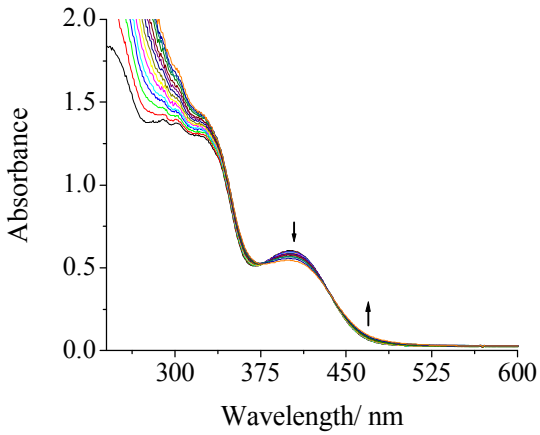


Fig. S3 Spectral changes obtained during titration of 20 mL of 3.3 × 10^{-5} M methanolic solution of $K_2[CH_2\{V^VO_2(sal\text{-}bhz)\}_2]\cdot 2H_2O$ **2** with dilute solution of 30% aqueous H_2O_2 (0.401 g, 3.01 mmol in 20 mL MeOH). Spectra were recorded after 25 min of every addition of dilute H_2O_2.

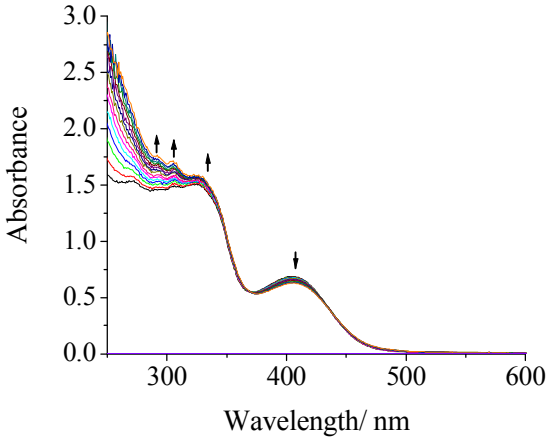


Fig. S4 Spectral changes obtained during titration of 20 mL of 4.7 × 10^{-5} M methanolic solution of $K_2[CH_2\{V^VO_2(sal\text{-}fah)\}_2]\cdot 2H_2O$ **5** with dilute solution of 30% aqueous H_2O_2 (0.401 g, 3.01 mmol in 20 mL MeOH). Spectra were recorded after 25 min of every addition of dilute H_2O_2.

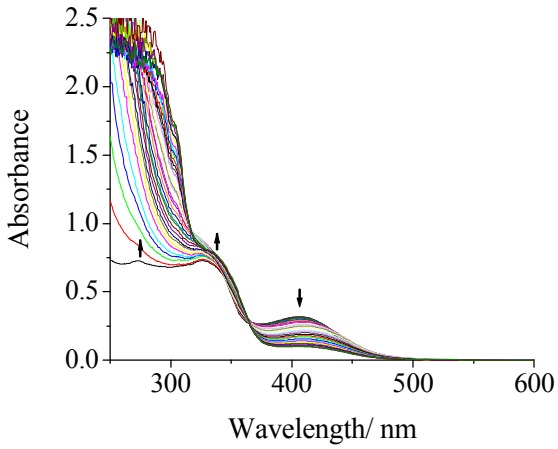


Fig. S5 Spectral changes obtained during titration of 20 mL of 3.3 × 10^{-5} M methanolic solution of $Cs_2[CH_2\{V^VO_2(sal\text{-}fah)\}_2]\cdot 2H_2O$ **6** with dilute solution of 30% aqueous H_2O_2 (0.401 g, 3.01 mmol in 20 mL MeOH). Spectra were recorded after 25 min of every addition of dilute H_2O_2.

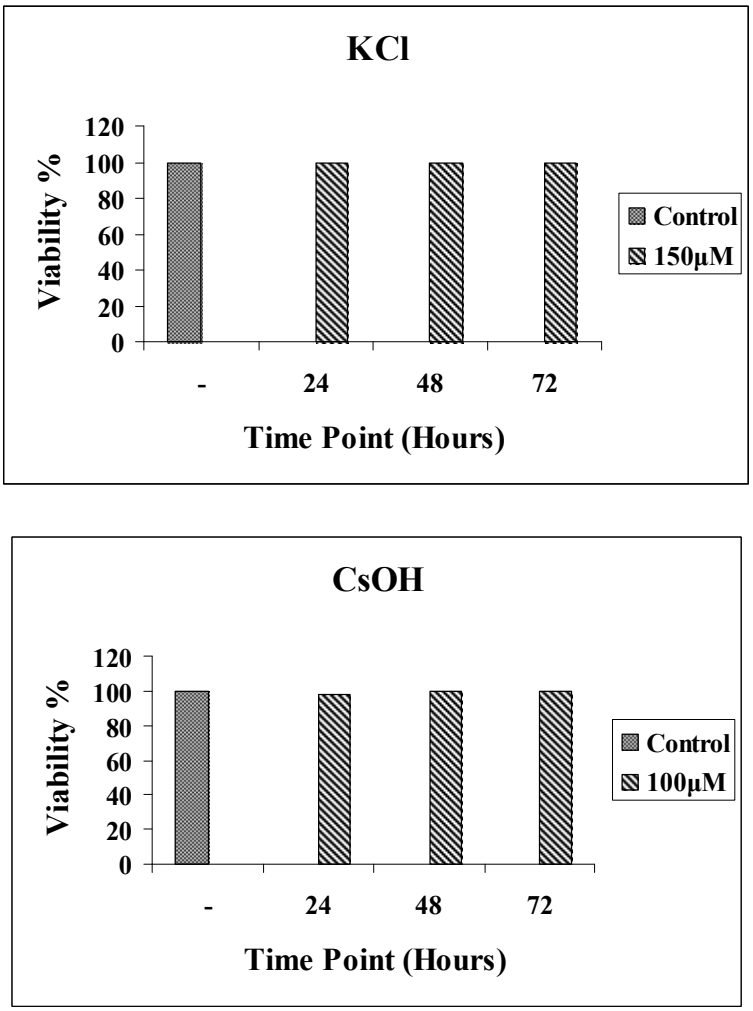


Fig. S6 Percentage of viable cells after 24 h, 48 h and 72 h on human cervical (HeLa) cells on incubation with KCl (150 µM, above) and CsOH (100 µM, below) or vehicale (DMSO). Cell survival was determined by MTT assay. ±SD bars are not visible as most of them are less than 0.04.