

# Highly Cytotoxic Vanadium(V) Complexes of Salan Ligands; Insights on the Role of Hydrolysis

## Supporting Information

Lilia Reytman,<sup>a</sup> Ori Braitbard<sup>b</sup> and Edit Y. Tshuva<sup>a,\*</sup>

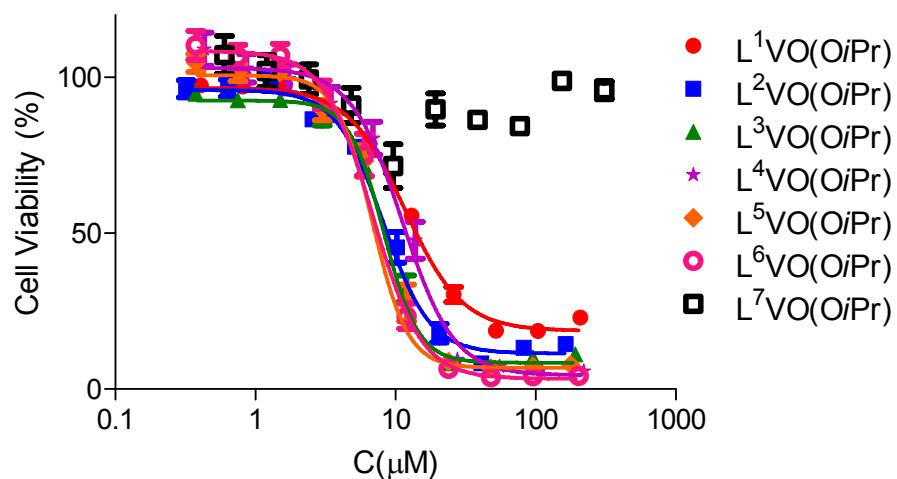
<sup>a</sup> Institute of Chemistry, The Hebrew University of Jerusalem, 91904, Jerusalem, Israel.

Fax: +972 2 6584282

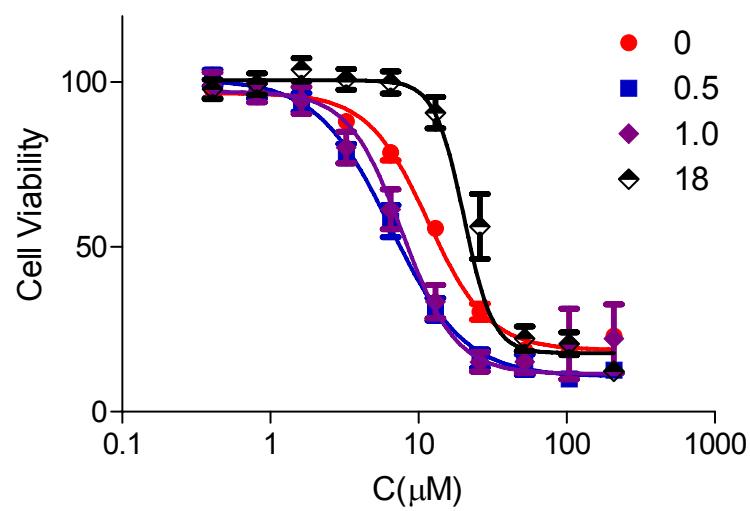
E-mail: [tshuva@chem.ch.huji.ac.il](mailto:tshuva@chem.ch.huji.ac.il)

<sup>b</sup> Department of Cell and Developmental Biology, Alexander Silberman Institute of Life Sciences, The Hebrew University of Jerusalem, 91904, Jerusalem, Israel.

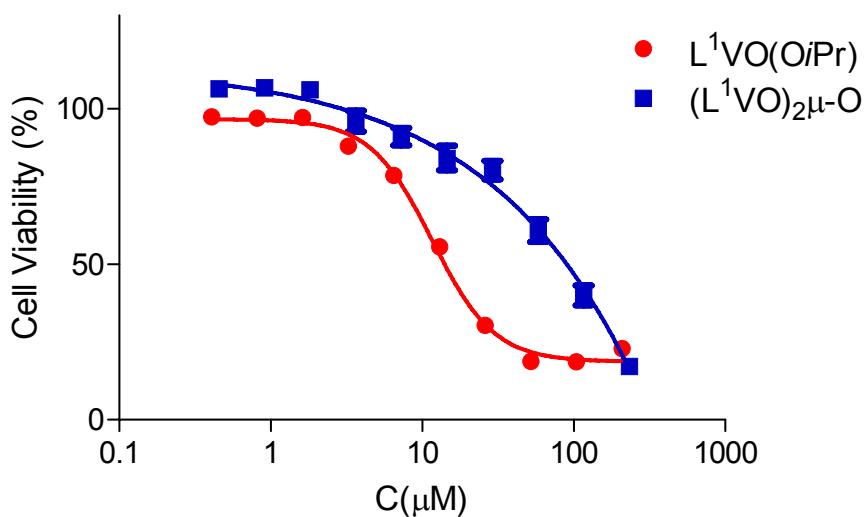
### Cytotoxicity Measurements on OVCAR-1 Cells



**Figure 1.** Dependence of OVCAR-1 cell viability on administered concentration of  $L^1$ - $^7$ VO(O*i*Pr) following 3 days incubation obtained by the MTT assay



**Figure 2.** Dependence of OVCAR-1 cell viability on administered concentration of  $L^1$ VO(O*i*Pr) following pre-incubation in aqueous medium for varying periods (given in hours) prior to cell addition and 3 days incubation with cells, obtained by MTT assay

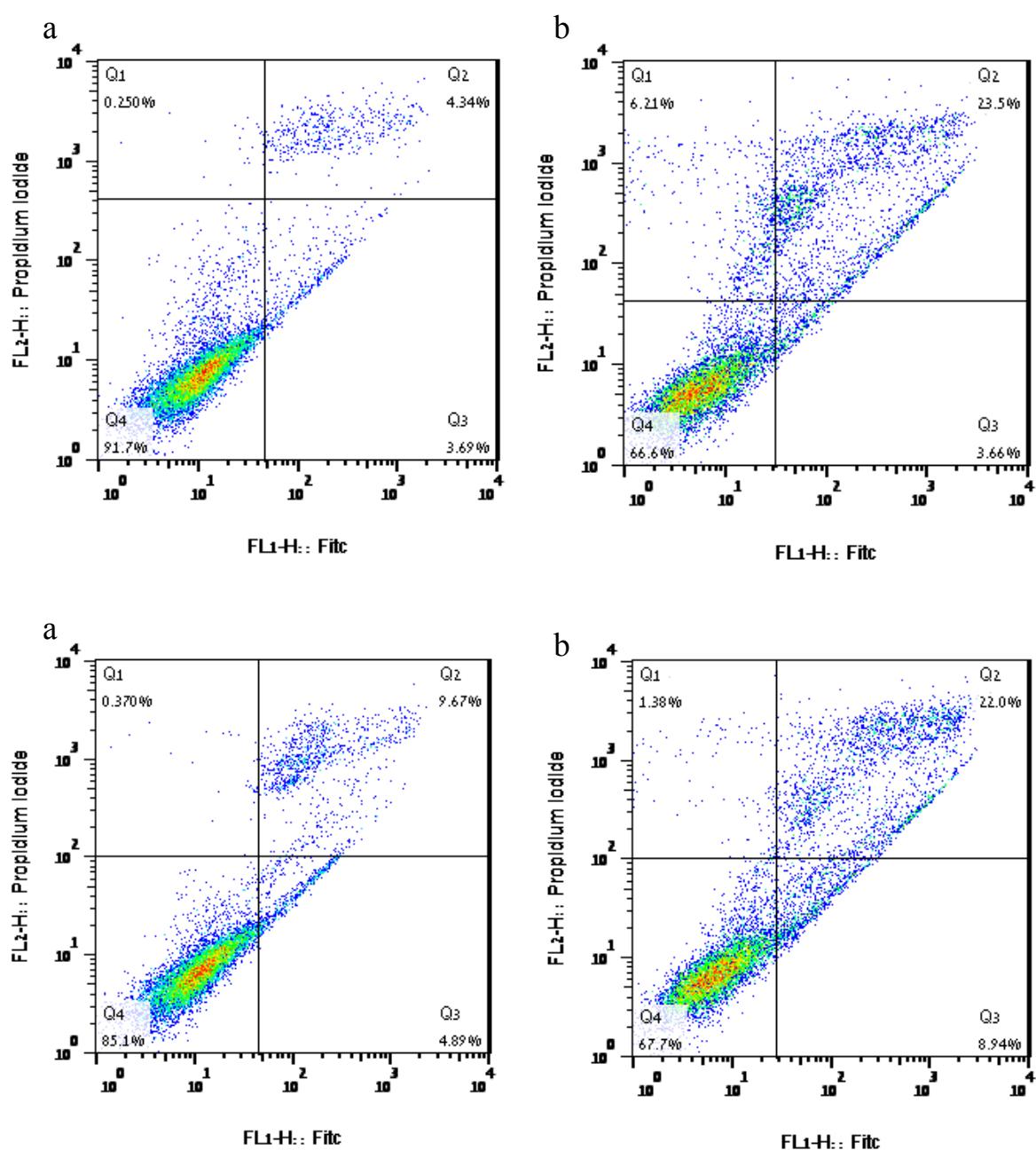


**Figure 3.** Dependence of OVCAR-1 cell viability on administered concentration of  $L^1VO(OiPr)$  and its hydrolysis product  $(L^1VO)_2\mu-O$  following 3 days incubation obtained by the MTT assay

### **Apoptosis vs. Necrosis Measurements**

Apoptosis was analyzed using the MEBCYTOR Apoptosis kit (MBL, Japan). 50,000 cells were seeded into 6 well plate. Following 24 hours, the cells were exposed to the compound and incubated for additional 24 hours. Cells were trypsinized and washed twice with PBS, resuspended in 85 µL of binding buffer and 10 µL of annexin V-FITC and 5 µL of propidium iodide were added. Cells were mixed well and incubated for 15 minutes in the dark. 400 µL of binding buffer was added and cellanalysis was performed on a Becton-Dickinson Excalibur Fluorescence Activated Cell Sorter using a single laser emitting excitation light at 488 nm. FloJo program was used for peak fluorescence quantification.

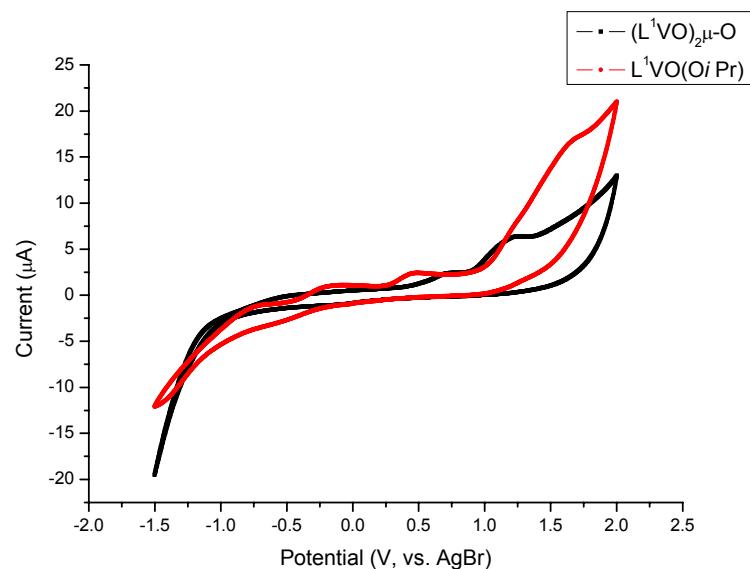
See Figure on the following page.



**Figure 4.** FACS analysis of apoptosis vs. necrosis showing propidium iodide against Annexin expression of HT-29 (top) and OVCAR-1 (bottom) cells that were (a) untreated or (b) treated with 100  $\mu$ M of  $L^1VO(OiPr)$  and incubated for 24 hours

### Cyclic Voltammetry Measurements

Cyclic voltammetry measurements were performed at room temperature on a μAutolab type III potentiostat and analyzed with GPES software. Tetrabutylammonium hexafluorophosphate (TBAPF<sub>6</sub>) (0.1 M), used as supporting electrolyte, was obtained from Sigma Inc. An electrochemical three-electrode cell was used with a platinum electrode as the working electrode, a platinum wire as the counter electrode and AgBr electrode as the reference electrode. The scan was performed at 0.2 v/s scan rate, in a THF solution containing 2 mM of the studied complexes.



**Figure 5.** Cyclic voltammetry of  $\text{L}^1\text{VO(OiPr)}$  and  $(\text{L}^1\text{VO})_2\mu\text{-O}$  in a THF solution