Supporting Online Material for

Cytotoxic effects of two organotin compounds and their mode of inflicting cell death on

four mammalian cancer cells

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Figure S1 and legend for Figure S1

Bis(Ph ₂ SnCl)Xylene Concentration	Values of Cytotoxicity	Average	Standard Deviation	Calculated CC ₅₀ *
50 µM	46.5	44.86667	3.003886	58.65
	46.7			
	41.4			
100 µM	74.5	74.53333	0.152753	
	74.4			
	74 7			

 $*CC_{50}$ in μ M is defined as the concentration of chemical compound necessary to disrupt the plasma membrane of 50% of the cell population, after 16 h of incubation. Cells with compromised plasma membranes were monitored with Propidium iodide (PI) and flow cytometry.

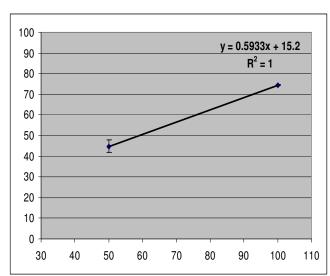


Figure S1. HeLa Cells treated with two different concentrations of Bis(Ph₂SnCl)Xylene. Representative values with their associated calculations (a) and graph with its corresponding equation (b) utilized to determine the CC_{50} . In this example, averages of triplicate values were plotted against chemical compound concentration as described in Material and Methods. The equation of the straight line of best fit was used to calculate the concentration of the chemical compound inflicting 50% of cell death.

b

а

Figure S2 and legend for Figure S2

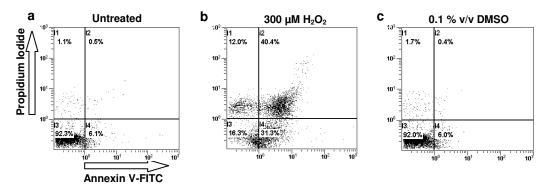


Figure S2. Representative flow cytometric histograms of controls for apoptosis/necrosis experiments using as an example NFS-70 pro-B murine lymphoblast cell line. Cells were untreated (a), treated with 300 μ M H₂O₂ (b), and treated with 0.1 μ l v/v of DMSO (c). Experiments with less than 90% of viability for untreated cells were discarded. Each histogram is divided in four quadrants; left top quadrant – Necrosis, cells that lost their membrane integrity permeable to propidium iodide, without FITC signal, one color (red); right top quadrant - late apoptosis, cells with compromised plasma membrane, permeable to propidium iodide, but also reactive to Annexin V-FITC, two colors (green and red); Lower left quadrant - alive unstained cells, without propidium iodide of FITC signal, no fluorescent signal; Lower right quadrant-early apoptosis, cells reactive to Annexin V-FITC, one color (green). Approximately 10,000 events were recorded and analyzed for each individual sample using CXP software (Beckman Coulter).