



**Cell Viability Assessment of Four Test Articles (MTT) in Human Kidney
Proximal Tubule Cells**

Cyprotex Study Number: CYP1751-R1A

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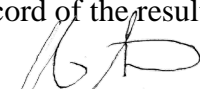
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AUTHENTICATION STATEMENT

I, the undersigned, hereby declare that the work described in this report was performed according to the study protocol and/or standard procedures, and to the best of my knowledge, this report provides a correct record of the results obtained.



Associate Director

October 31, 2017

Date

1 PURPOSE

The objective of this study was to assess the cytotoxicity potential of four test articles, AB070597 and in Human kidney proximal tubule cells

2 STUDY CONDITIONS

This study was performed under non-GLP conditions. All work was performed with appropriate local health regulations and ethical approval.

3 EXPERIMENTAL DESIGN

3.1 Test Articles

Test Article	Actual MW (FW)
	354.31
	164.20
AB070597	n/a
	226.2

3.2 Cell Viability: experimental conditions

Test Article	Test conc.	Reference compound
	500, 250, 125, 62.5, 31.25, 15.63, 7.81, 3.91, 1.95 and 0.98 μ M	CCCP
	500, 250, 125, 62.5, 31.25, 15.63, 7.81, 3.91, 1.95 and 0.98 μ M	CCCP
AB070597	100.00, 50.00, 25.00, 12.50, 6.25, 3.13, 1.56, 0.78, 0.39 and 0.20 μ g/ml	CCCP
	500, 250, 125, 62.5, 31.25, 15.63, 7.81, 3.91, 1.95 and 0.98 μ M	CCCP

Experimental Procedure

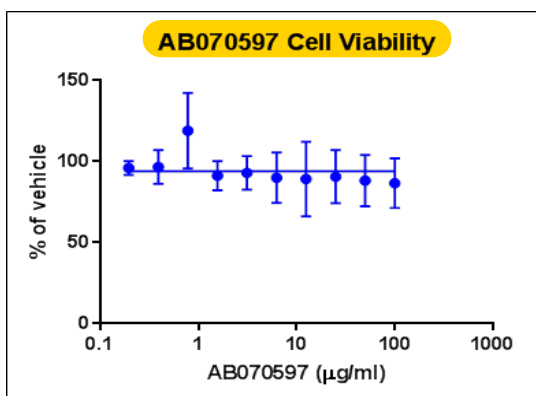
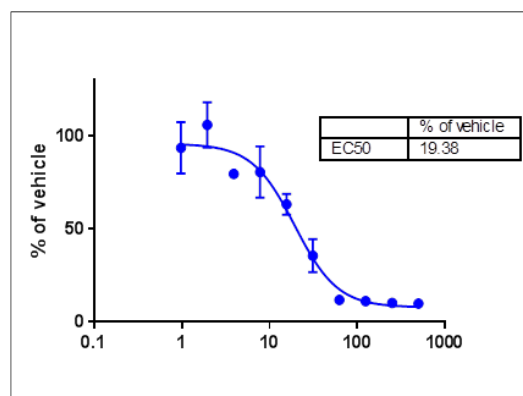
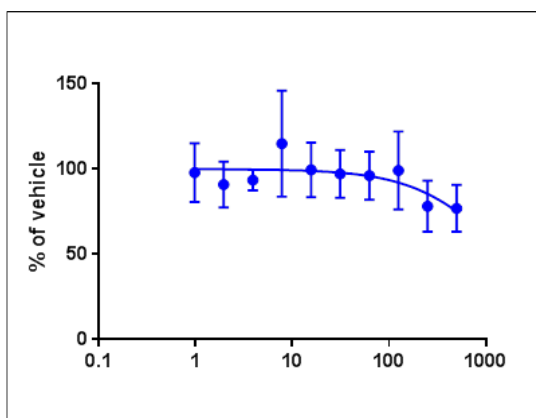
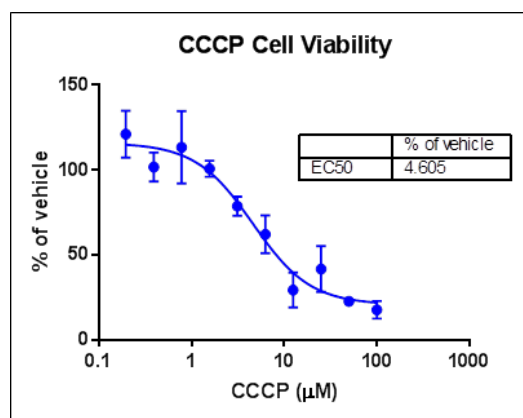
Human kidney proximal tubule cells were plated on 384-well tissue culture treated polystyrene plates at 1.5×10^4 cells. After an overnight incubation at 37°C, the cells were dosed with test compounds and controls at a range of concentrations and incubated for 72 hr at 37°C. Cell viability was measured using the Promega CellTiter 96 Non-Radioactive Cell Proliferation Assay (MTT) kit by adding the Dye Solution to each well and incubating for 3 hr at 37°C. After incubation, the Solubilization Solution/Stop Mix was added to each well. Plates were incubated at 37°C for 1 hr, mixed on a plate shaker for 10 min and then absorbance was read at 570nm. The EC₅₀ was calculated using GraphPad Prism.

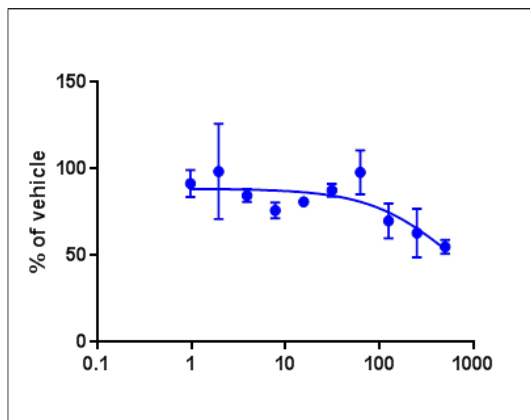
4 RESULTS AND CONCLUSIONS

4.1 Cell Viability: Summary

Test Article	Cell Viability IC ₅₀	Comment
CCCP	4.605 μ M	Control
	> 500 μ M	Test Article
	19.36 μ M	Test Article
AB070597	> 500 μ g/ml	Test Article
	546.9 μ M	Test Article

4.2 Cell Viability: Individual Data





4.3 Conclusions

The compounds were tested in Human Kidney Proximal Tubule cells. The positive control compound responded as expected. and AB070597 were not cytotoxic up to their top concentration tested.