In vitro effects of 3-(4-Dimethylamino-naphthelen-1-ylmethylene)-1, 3-hydroindolo-2-one) in combination with epigallocatechin gallate on tumour survival in melanoma cells

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Introduction

Cancer can be described as the growth and division of cells without regulatory systems. The international agency for cancer reported 10 million deaths worldwide, 1 million of those in Africa and a quarter of those from South Africa [1]. These statistics reinforce the rising prevalence of deaths attributed to cancer and emphasizes the need for research focusing on targeting specific types of cancers. Melanoma is a cancer of the skin melanocytes which has an aggressive malignancy and a survival rate of less tha 5 years [2] . Current therapeutics for melanoma are limited in either efficacy or safety Therefore, this project will focus on determining the possible anticancer ability of alternative therapeutics such as phytochemicals EGCG and anticancer compound MAZ-51.

Aims & Methods

The aim of this study was to determine the efficacy of MAZ-51 and EGCG on tumour cell survival. In this study the *in vitro* effects of MAZ-51 and EGCG on tumour cell survival was determined using B16F10 melanoma and RAW 264.7 murine macrophage cell lines. The following methods were used:

Crystal violet assay (CV) : To determine the percentage cell viability and IC_{50} values

PlasDIC Microscopy : To determine morphological changes at IC_{50} with a pseudo 3D effect

Light microscopy Haematoxylin & Eosin (H&E) staining : To determine morphological changes at IC_{50}

Results & Discussion

The most significant IC_{50} values were obtained at 72 hours, 109 µM for EGCG and 128.7 µM for MAZ-51. Both compounds had significant p values < 0.0001 at the highest concentrations (Figure 1). Conversely, RAW 264.7 cells IC_{50}'s were much higher, 6,384x10^5 µM for EGCG and 811,8 µM for MAZ-51, with no statistical significance (Figure 1). Morphological changes at IC_{50} concentrations showed decreased cell density, cell rounding and blebbing indicative of apoptosis, with RAW 264.7 cells having less of an effect (Figure 2 &3). Future studies can combine EGCG and MAZ-51 IC_{50}'s to determine if it will yield a stronger additive effect.

Conclusion

In conclusion both MAZ-51 and EGCG showed a reduction in tumour cell growth. The results of this study is in agreement with literature thus far, as each compound has demonstrated a significant reduction in tumour cell growth. Therefore, making EGCG and MAZ-651 are ideal candidates for alternative therapeutic agents against melanoma

References

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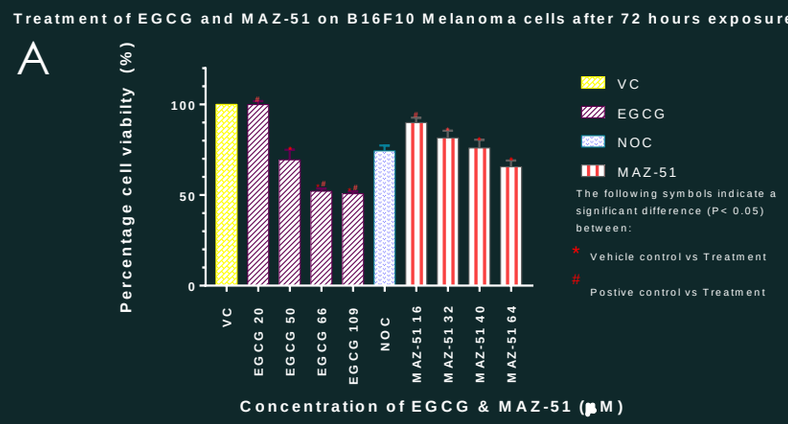
CV assay Results IC_{50}



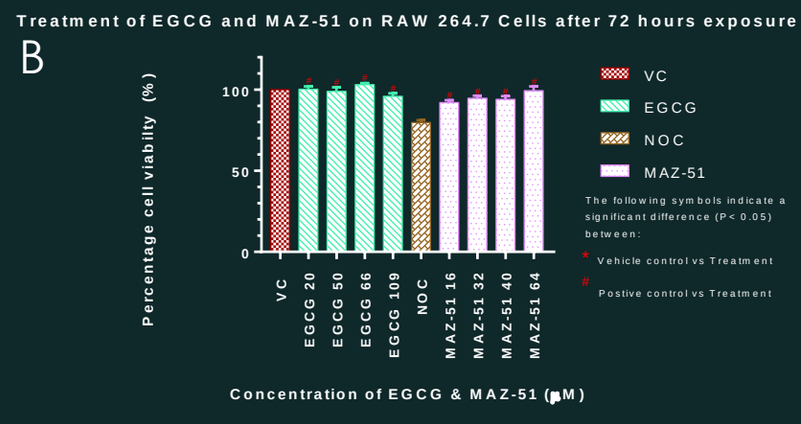


Figure 1A & B: Crystal violet cell viability analysis of B16F10 and RAW 264.7 cell respectively, using GraphPad Prism v6.01 (California, USA).

PlasDIC Results at IC_{50}

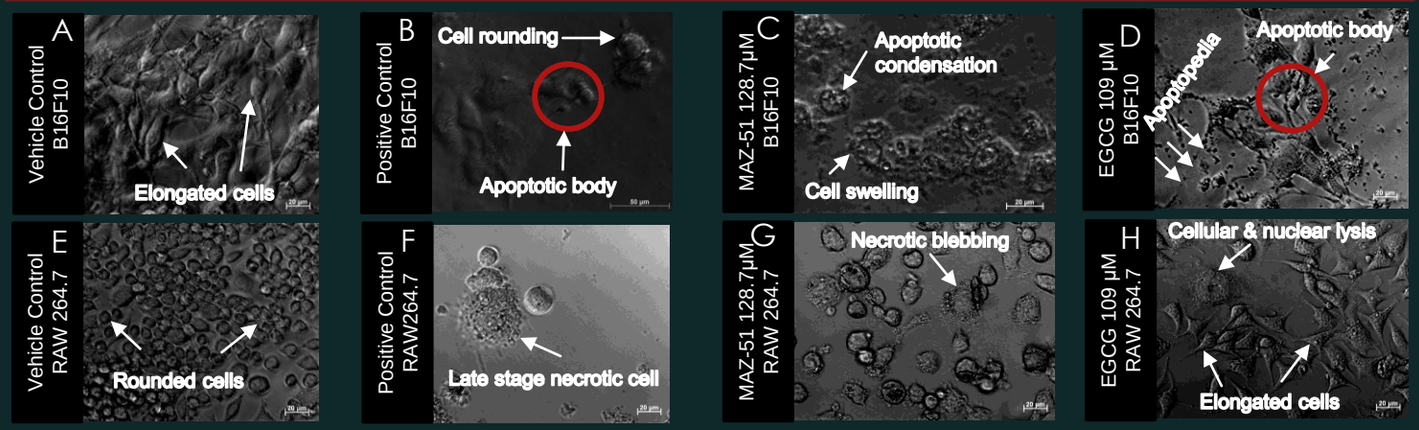


Figure 2: Pseudo 3D Morphological analysis of cells exposed at IC_{50} of MAZ-51 and EGCG at 40X magnification. A-D B16F10 Cells, E-H RAW 264.7 Cells

H & E Results at IC_{50}

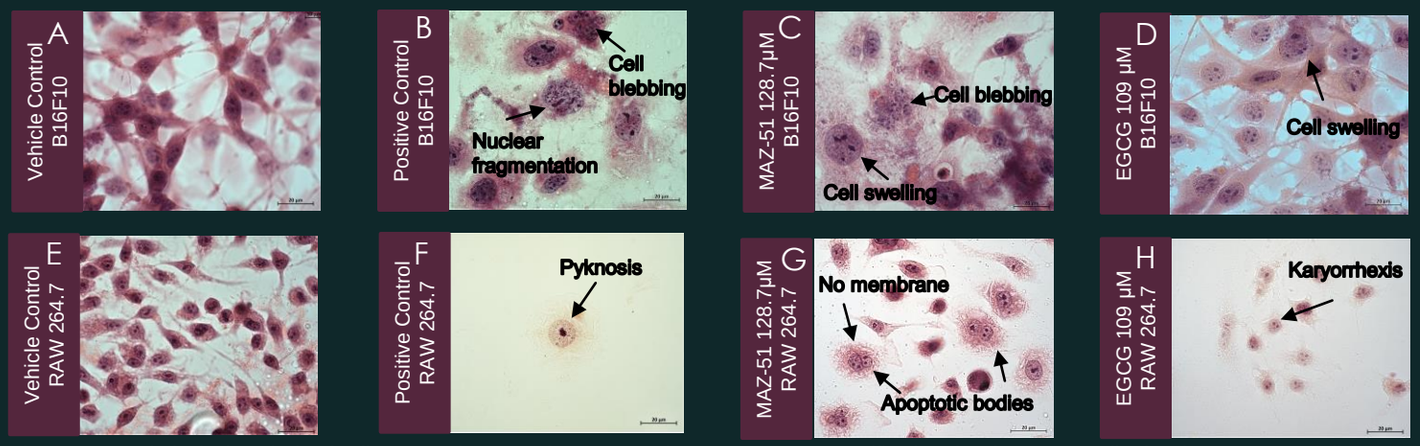


Figure 3: Morphological analysis of cells exposed at IC_{50} of MAZ-51 and EGCG at 100X magnification. (A-D) B16F10 Cells, (E-H) RAW 264.7 Cells using, Zeiss Axio Imager M2 light microscope (Germany).