Dose-dependent Response to Insulin and Bitter Melon Extract in MCF7 Cells Cultured in Low/Normal Glucose

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INTRODUCTION

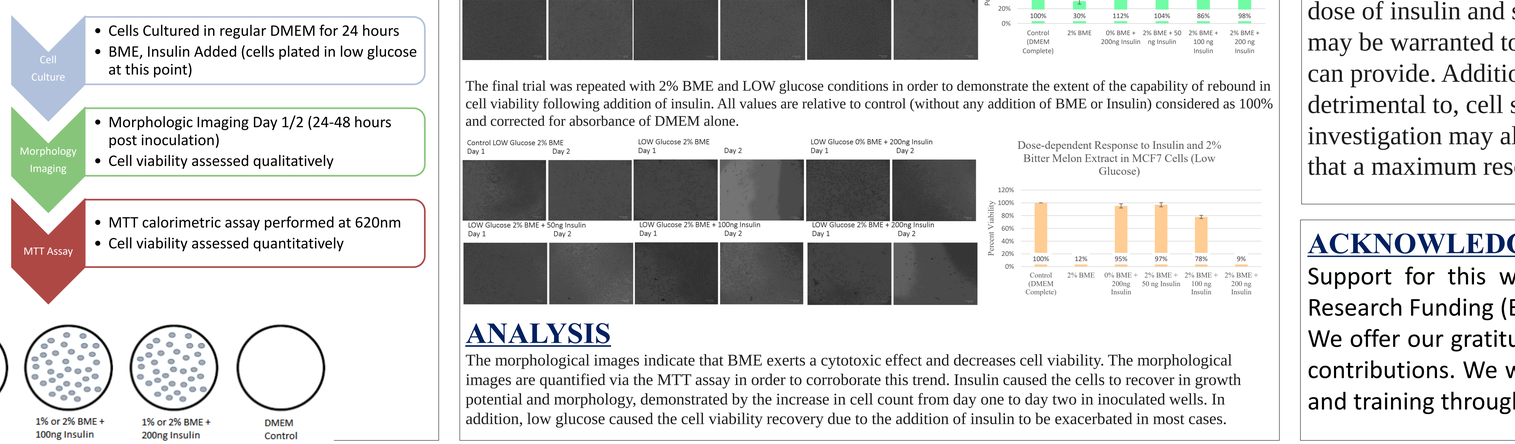
Bitter melon extract (BME) is known to inhibit breast cancer cells (MCF-7) proliferation. Due to the high metabolic rate of cancer cells, we hypothesized that low glucose levels are detrimental to cell survivability and that BME exerts a cytotoxic effect by altering the capability of a cell to utilize glucose. Through this study, we seek to investigate if insulin is capable of mitigating the cytotoxic effects of BME and provide a "rescue effect" that increases cell viability, or even completely reverses the cytotoxic effects of BME.

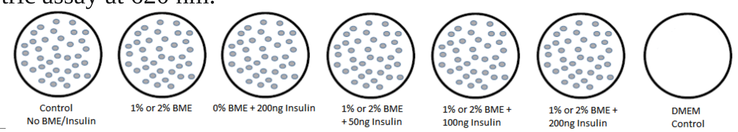
OBJECTIVE

In this study, we aim to test the effects of BME on MCF-7 cell (human metastatic breast cancer cell) survival and whether insulin is able to mitigate or counteract the cytotoxic effects of BME. We also repeated the trials using low-glucose medium in order to exacerbate the cytotoxic effects of BME and demonstrate the potential of the rescue effect of insulin.

METHODS

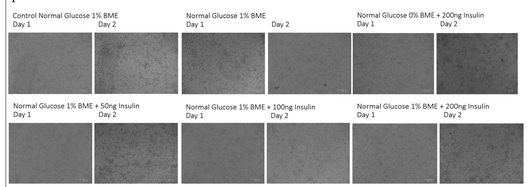
MCF-7 cells were cultured in DMEM medium with either 1% or 2 % BME (v/v) and in the presence or absence of insulin at various doses [50ng/mL, 100ng/mL, 200ng/mL]. This same experiment was repeated at 2% BME (v/v), however, cells were plated with DMEM low glucose medium (1g/L). Morphologic images were taken every 24 hours for 2 days. Cell viability was then assessed through an MTT colorimetric assay at 620 nm.

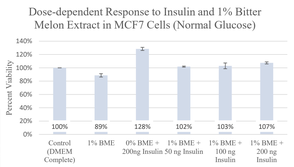




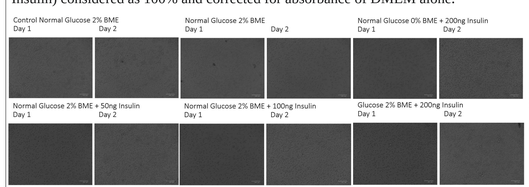
RESULTS

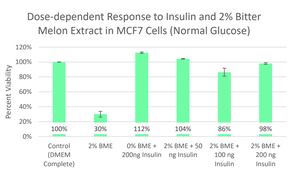
BME dose-dependently inhibited MCF-7 cell viability. Insulin demonstrated a rescue effect and increased the cell viability despite the addition of BME. Insulin also dose-dependently enhanced MCF-7 cell viability to an extent. Following were the % absorbance values with increasing doses of insulin. All values are relative to control (without any addition of BME or Insulin) considered as 100% and corrected for absorbance of DMEM alone. The 1% BME (bitter melon extract) is the positive control in our studies as our lab have found BME to be toxic to MCF-7 cells.





The same trial was repeated with 2% BME in order to better visualize the cytotoxic effects of BME and further demonstrate the subsequent rebound in cell viability following addition of insulin. All values are relative to control (without any addition of BME or Insulin) considered as 100% and corrected for absorbance of DMEM alone.





The final trial was repeated with 2% BME and LOW glucose conditions in order to demonstrate the extent of the capability of rebound in cell viability following addition of insulin. All values are relative to control (without any addition of BME or Insulin) considered as 100% and corrected for absorbance of DMEM alone.

ANALYSIS

The morphological images indicate that BME exerts a cytotoxic effect and decreases cell viability. The morphological images are quantified via the MTT assay in order to corroborate this trend. Insulin caused the cells to recover in growth potential and morphology, demonstrated by the increase in cell count from day one to day two in inoculated wells. In addition, low glucose caused the cell viability recovery due to the addition of insulin to be exacerbated in most cases.

CONCLUSIONS

The data indicates the addition of insulin is capable of rescuing cells from the cytotoxic effects of BME. Insulin seems to be capable of mitigating the cytotoxic effects of BME and, in some cases, even enhancing the viability of cells despite the influence of BME. However, in the low glucose experiments, the low cell viability observed in the 2% BME + 200ng Insulin warrants further consideration. The relatively diminished rescue effect with 200ng of insulin and 2% BME may be due to the inherent strain the cells were already experiencing with BME.

ONGOING/FURTHER RESEARCH

The high dose of insulin, coupled with the strain of attempting to replicate in sub-optimal conditions, may have led to cells that were incapable of meeting the metabolic demand placed by the large dose of insulin and subsequent influx of glucose. Further study may be warranted to discover the threshold of benefit that insulin can provide. Additionally, the amount of glucose necessary for, or detrimental to, cell survival may also be worth further study. Such investigation may allow for repeated trials with altered doses such that a maximum rescue effect and subsequent recovery is seen.

ACKNOWLEDGEMENTS

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