Cytotoxic and apoptotic effects of heat killed *Mycobacterium indicus pranii* (MIP) on cancer cell lines

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INTRODUCTION

Cancer is a leading cause of death worldwide, accounting for 8.2 million deaths in 2012. According to a report by the World Health Organization (WHO), it is expected that annual cancer cases will rise from 14 million in 2012 to 22 within the next 2 decades. So, improved therapeutic strategy is urgently needed. The idea of employing bacteria in cancer treatment arose in the early 1980s. *Mycobacterium indicus pranii* (MIP) is a non-pathogenic mycobacterium which is commercially available as a heat killed vaccine for leprosy and tuberculosis. Its immune-potentiating ability is an attractive property to utilize this bacterium in cancer treatment. It has been tested on several cancer types such as lung and bladder cancers where tumour regression and complete recovery was observed. Bacteria based anti-tumour therapy possess several advantages over chemical based drug1

- Firstly, some bacteria are able to selectively replicate and accumulate within tumour due to hypoxia environment and inhibits tumour growth.
- Motile bacteria are able to spread throughout the tumour and help in targeting systemic diseases.
- Bacteria can readily express multiple therapeutic transgenes such as cytokines and pro-drug converting enzymes to eradicate tumour mass.

METHOD

**Cancer cells**

- Plated in 96-well plates

**Treatment**

- Heat killed bacteria
- Heat killed supernatant
- Live bacteria
- Live supernatant

**MTT assay**

**PARP assay**

**DNA fragmentation assay**

RESULTS

**Cytotoxic screening for active MIP fractions**

**In vitro cytotoxic effects of MIP HKB**

**DISCUSSION**

- Four different fractions can be obtained from MIP: LB, HKB, LS and HKS, the most widely used fractions are HKB and LS.
- Among 4 fractions, only MIP HKB showed cytotoxic effects where cell viability reduced to 24% in CaSki and 26% in A549, while the remaining fractions did not show any killing effects.
- MIP HKB selectivity in MCF-7 and ORL-115 with IC50 values XXX µl/ml/106 cells, are lower than normal cells.
- According to a previous study on apoptotic cell death in *in vitro* by Pandey et al., (2011), 60-70 µl of MIP is required to induce cell death in 40-45% mouse peritoneal macrophages while in this study 100 µl MIP HKB induced 75% cell death at 8 hr.
- Cancer cell death was induced via apoptosis in MCF-7 and ORL-115 cells as confirmed through PARP and DNA fragmentation assays

CONCLUSION

- The HKB MIP was identified as the most potential cytotoxic fraction compared to LB, LS and HKS in terms of its low IC50 values and induction of apoptotic cell death in breast and oral cancer cells.
- Therefore, the cytotoxic and apoptotic effects of MIP heat killed bacteria in these two human cancer cells indicate that it can be a good candidate for further pharmacological studies to develop an effective anti-cancer agents.

REFERENCES


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Bacteria based anti-tumour therapy possess several advantages over chemical based drug1
- Firstly, some bacteria are able to selectively replicate and accumulate within tumour due to hypoxia environment and inhibits tumour growth.
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METHOD

**Cytotoxic screening for active MIP fractions**

**In vitro cytotoxic effects of MIP HKB**

**RESULTS**

**DISCUSSION**

- Four different fractions can be obtained from MIP: LB, HKB, LS, and HKS, the most widely used fractions are HKB3 and LS3.
- Among 4 fractions, only MIP HKB showed cytotoxic effects where cell viability reduced to 24% in CaSkis and 26% in A549, while the remaining fractions did not show any killing effects.
- MIP HKB selectivity in MDA-MB-231, MCF-7, CaSkis, A549, SK-LU-1, DU-145, HepG2, ORL-48, ORL-115 and ORL-136 with IC50 values between 5.6 to 21 μg/ml/106 cells, all of which are lower than normal cells.
- According to a previous study on apoptotic cell death in *in vitro* by Pandey et al., (2011), 60-70 μl of MIP is required to induce cell death in 40-45% mouse peritoneal macrophages while in this study 100 μl MIP HKB induced 75% cell death at 8 hr.
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