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Anticancer activity of *Trichoderma harzianum* extract against NCI-H292 lung cancer cells

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Cancer is one of the leading causes of death worldwide. Chemotherapy has been the choice of cancer treatment for many years however, it can also affect normal cells and create many undesirable side effects and have the potential to develop resistance. Therefore, investigators must reassess their approach to translate discovery research into greater clinical success and impact aiming to find novel compounds. Endolichenic fungi (ELF) are potential source of producing many bioactive compounds. Preparations of ELFs extracts are commonly used to search for anticancer activity. Based on the fact that fungal extracts provide evidence to develop anticancer drugs, this study was conducted to evaluate the anticancer activity of an ELF, *Trichoderma harzianum,* (strain No: MF029755) extract against NCI-H292 lung cancer cells. Organ specific *in-vitro* assays are imperative in large scale screening of natural products with useful clinical activity. Among many such assays, sulforhodamine B (SRB) assay employs a protein binding aminoxanthene dye, to provide a quantitative analysis of viable cells in a culture following the introduction of the compound. Preliminary investigations revealed that crude ethylacetate extract of an endolichenic fungus, *T. harzianum,* and chloroform fractions of crude extract (12.5 mgL^{-1}, 25 mgL^{-1}, 50 mgL^{-1}, 100 mgL^{-1} and 200 mgL^{-1}) obtained by partition were positive for the SRB assay. IC_{50} values of crude extract and the chloroform fraction were 68.48 mgL^{-1} and 38.44 mgL^{-1} respectively. The chloroform fraction was chromatographed over silica gel column to obtain seven fractions. Cytotoxicity of the seven fractions obtained from the crude extract of the fungus was determined using SRB assay against lung cancer cell line NCI-H292 following standard protocols. The cell suspension in Dulbecco's Modified Eagle Medium (DMEM) was aliquoted into 96-well plate. After incubation cells were treated with two concentrations (100 mgL^{-1} and 200 mgL^{-1}) of fractions obtained by column chromatography. SRB dye was added to each well and acetic acid was used to remove unbound dye. Absorbance was measured at 540 nm using microplate reader. Survival percentage of the cells was calculated. If no viable cells present pink color of the medium turns colorless. In the current assay control wells and 1^{st} fraction remained pink and all the other treatments turned pink into colorless. Seventh fraction showed the highest activity and further purification, SRB assays and structure elucidation will be carried out.

Keywords: Cytotoxicity, SRB assay, *Trichoderma harzianum*