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Conference Presentation

In vitro studies of Psorinum 6x on several human cancer cell lines reveal its anti-cancer potential

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Abstract

Objective: Psorinum therapy is claimed to combat/ameliorate a variety of human cancers, but for obvious reasons these studies are devoid of any untreated/placebo-treated controls. Therefore, if *Psorinum* 6x administration to cancer cells of different origin can show visible anti-cancer effects in a controlled *in vitro* study has been examined.

Materials & Methods: *Psorinum* 6x was provided by Hahnemann Publishing Company (HAPCO), 165 BB Ganguly Street, Kolkata for our research. It was prepared by HAPCO following standard homoeopathic guidelines from authentic pus cells obtained from an eczema patient being treated at National Institute of Homeopathy, Salt Lake, Kolkata. MTT assay was initially done on several cancer cell lines like A549 (lung cancer), HeLa (cervix cancer), HepG2 (liver cancer) and MCF7 (breast cancer). *Psorinum* 6x showed strongest anticancer effect against A549 though it also had lesser effect against other cell lines tested. Therefore, A549 was chosen as the model cell line for further study using several relevant protocols. Effects of *Psorinum* 6x were compared with that of "Placebo 6x" control made of the same stock of "vehicle" used for preparation of *Psorinum* 6x. Protocols like analysis of cell cycle progression, generation of reactive oxygen species (ROS), change in mitochondrial membrane potential (MMP) and actual cell death (apoptosis), if any, were analysed flow-cytometrically. Whether *Psorinum* 6x could damage DNA and induce morphological changes were also determined microscopically. Expression of different signal proteins related to cell death (apoptosis) and survival were critically studied by western blot analysis and confocal microscopy. Further, to determine if *Psorinum* 6x could interact directly with DNA to induce any conformational changes was also determined by circular dichroism (CD) spectroscopy.

Results: *Psorinum* 6x treatment reduced cell viability and inhibited cell proliferation at 24h after treatment, arresting cell cycle at sub-G stage. Upon *Psorinum* treatment, there were increase of ROS and MMP depolarization, morphological changes and DNA damage typical of apoptosis in A549 cells, along with externalization of phosphatidyl serine. Further, an increase in p53 level, Bax translocation into mitochondria, cytochrome c release into cytosol along with reduction of Bcl2 level and caspase 3 activation were noted which eventually drove A549 cells towards apoptosis; the apoptotic signalling was found to be through mitochondria-mediated caspase 3 dependent

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pathway. Evidence of direct interaction of *Psorinum* with cellular DNA was revealed from CD-spectroscopy.

Conclusion: Thus, *Psorinum* 6 x had positive anti-cancer effects against a number of cancer cells, of which it appeared to have strongest effect against the lung cancer cell, A549.

Keywords: Psorinum therapy, reactive oxygen species, anticancer potential, apoptosis, drug-DNA interaction.

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