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Caspase 3/7 activation during apoptotic cell death of human Rhabdomyosarcoma (RMS) and breast adenocarcinoma (MCF-7) cells induced by different fractions of *Chnoospora minima*

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Marine seaweeds are a rich source of bioactive metabolites that can be used as a source for the development of anti-cancer drugs. Apoptosis is a form of programmed cell death involved with the elimination of unwanted cells from the body. Among different mechanisms of apoptosis, caspases are a family of protease enzymes playing an essential role in apoptosis. Therefore, the present study was aimed to determine the caspase 3/7 activation in human rhabdomyosarcoma (RMS) and breast adenocarcinoma (MCF-7) cells following treatment with hexane and chloroform fractions of the seaweed species *Chnoospora minima*. The apo-one homogenous caspase 3/7 activity of treated cells was evaluated according to the manufacturer`s instructions (G7790, Promega, USA). Polysaccharide depleted polyphenol-rich methanol extract was sequentially partitioned with hexane, chloroform, and ethyl acetate to determine the cytotoxic activity. Based on the results, hexane and chloroform fractions of *C.minima* were selected to determine the caspase 3/7 activation of human RMS and MCF-7 cells. The caspase 3/7 activation was quantified by relative flurescence unit (RFU). The chloroform fraction ($RFU_{4\ hrs}$:3932.9) of *C.minima* showed prominent activation of caspase 3/7 in RMS cells after 4 h of caspase treatment more than the hexane fraction ($RFU_{4\ hrs}$:2556.6) compared to the standard Staurosporine ($RFU_{4\ hrs}$:3417.5) and cycloheximide ($RFU_{4\ hrs}$:2950.5). In contrast, hexane ($RFU_{3\ hrs}$:1496.9) and chloroform ($RFU_{3\ hrs}$:1464.7) fractions treated MCF-7 cells showed low caspase 3/7 activation, and the highest activity was observed after 3 h of caspase treatment. Hence, it can be concluded that the hexane and chloroform fractions of *C.minima* induce apoptosis in RMS cells more prominently via the caspase 3/7 pathway compared to the MCF-7 cells. Therefore, further studies should be conducted to confirm the activity of caspase 3 and 7 via gene expression analysis.

Keywords: Anti-cancer, RMS, MCF-7, *Chnoospora minima*, Caspase 3/7

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