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Evaluation of *paavu chooranam* prescribed to cure breast cancer by the *cheruthikonam* traditional siddha medicinal practitioner of Kanyakumari district, India

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Breast cancer is the leading cause of death in women worldwide among other types of cancers. Paavu chooranam is a siddha polyherbal formulation prescribed to cure breast cancer patients was prepared by using the ingredients kottam and venthayam (1 Kalanchi), illavangam, venkadugu, vasambu and amukkara (2 kalanchi), chukku, milagu, thippili, athimathuram, omam, kadugu rohini and kandankatthiri (3 kalanchi), nellikai, kadukkai, thandrikkai, jathikai, jathipathiri, vaalmilagu and indhuppu (4 kalanchi), seeragam, karamjiragam, seenthil, adathodaiver and thoothuvalai (5 kalanchi). The present investigation was mainly focused on the scientific analysis of qualitative, quantitative, antioxidant, cytotoxic and apoptotic activities of paavu chooranam. The phytochemical constituents in the herbal formulation revealed positive response of significant secondary metabolites. The unexplored area of paavu chooranam towards their antioxidation effect of hydroxyl radical scavenging, DPPH, nitric oxide radical scavenging, hydrogen per oxide radical scavenging and reducing power activity in aqueous, silver nitrate and ethanol extracts indicated promising antioxidant activities in a dose dependent manner. The cytotoxic effect of control L929 fibroblast cell line exhibited 100% inhibition. The LC50 value (ED50 plus software V1.0) was obtained at 64.71 µg per mg. The morphological detection of apoptotic and necrotic cells determined by acridine orange and ethidium bromide double staining of control cells observed under fluorescent microscope showed the presence of living cells (normal green nucleus) and absence of early apoptotic, late apotoptotic and necrotic cells but paavu chooranam (medicine) showed maximum detection of necrotic cells (uniformly orange stained cell nuclei), followed by early apoptotic (bright green nucleus with condensed or fragmented chromatin), late apoptotic (orange stained nuclei with chromatin condensation or fragmentation) and living cells (normal green nucleus). Determination of apoptosis by flow cytometry revealed that the concentration of control profile 1.45E+06 cells/ml gated 94.74% of live cells (LL), early apoptotic (LR) 3.99E+03 cells/ml gated 0.26%; late apoptotic/Dead (UR) 4.51E+04 cells/ml gated 2.94%; debris (UL) 3.16E+04 gated 2.06% and late apoptotic/dead, debris 4.91E+04% cells/ml gated 3.20%. On the other hand, apoptosis profile of paavu chooranam showed that the concentration of 1.13E+06 cells/ml gated 77.93% live cells (LL), early apoptotic (LR) 2.83E+05 cells/ml gated 19.54%; late apoptotic/Dead (UR) 3.65E+04 cells/ml gated 2.52%; debris (UL) 1.45E+02 and late apoptotic/dead, debris 0.01% cells/ml gated 0.01%. The scientific analysis of siddha breast cancer medicines and case reports highlight the effect of medicine to relapse from cancer. This research work makes the society to believe that treatment is also possible without any significant side effects. Apparently, the promising active principles and underlying mechanism by which this activity was exhibited need to be further investigated.

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