

***Ruellia tuberosa* Ethyl Acetate Leaf Extract Induces Apoptosis and Cell Cycle Arrest
in Human Breast Cancer Cell Line, MCF-7**

ABSTRACT

Ruellia tuberosa L. has been previously shown to possess antioxidant and antiproliferative activities on cancer cells but its underlying mechanisms are largely unknown. This study aimed to elucidate the mode of action underlying this inhibitory effect on MCF-7 using ethyl acetate extract obtained after liquid-liquid partition of methanol crude extract. Antiproliferative effect of *R. tuberosa* ethyl acetate leaf extract (RTEAL) was evaluated using MTT assay. Its ability to induce apoptosis was assessed by DNA ladder formation, JC-1, Annexin V, and methylene blue staining assays. Perturbation of cell cycle progression was determined using flow cytometry. RTEAL was found to selectively inhibit the proliferation of MCF-7 cells with the IC₅₀ value of 28 µg/mL. Morphological changes such as nuclear fragmentation and chromatin condensation were observed although DNA laddering was undetected in agarose gel. RTEAL-induced apoptotic pathways by inhibiting the expression of anti-apoptotic BCL-2 while upregulating pro-apoptotic BAX, caspase 7 and caspase 8. RTEAL also caused cell cycle arrests at the S and G2/M phase and dysregulation of cell cycle regulators. These findings collectively demonstrate that RTEAL extract inhibited cell growth by inducing apoptosis and cell cycle arrest, suggesting its therapeutic potential against breast cancer.