

## **High-throughput metabolomics reveals dysregulation of hydrophobic metabolomes in cancer cell lines by *Eleusine indica***

### **ABSTRACT**

*Eleusine indica*, which is used in traditional medicine, exhibits antiproliferative activity against several cancer cell lines. However, metabolomic studies to evaluate the metabolite changes induced by *E. indica* in cancer cells are still lacking. The present study investigated the anticancer effects of a root fraction of *E. indica* (R-S5-C1-H1) on H1299, MCF-7, and SK-HEP-1 cell lines and analyzed metabolic changes in the treated cancer cells using ultra-high-performance liquid chromatography high-resolution mass spectrometry (UHPLC-HRMS). Cell metabolic activity assays demonstrated that the cell viability of the three cancer cell lines was significantly reduced following treatment with R-S5-C1-H1, with half-maximal inhibitory concentrations values of 12.95  $\mu\text{g/mL}$ , 15.99  $\mu\text{g/mL}$ , and 13.69  $\mu\text{g/mL}$  at 72 h, respectively. Microscopy analysis using Hoechst 33342 and Annexin V fluorescent dyes revealed that cells treated with R-S5-C1-H1 underwent apoptotic cell death, while chemometric analysis suggested that apoptosis was triggered 48 h after treatment with R-S5-C1-H1. Deconvoluted cellular metabolomics revealed that hydrophobic metabolites were significantly altered, including triacylglycerols, phosphatidylcholine, phosphatidylethanolamine, sphingomyelin, and ceramide, suggesting that apoptosis induction by R-S5-C1-H1 potentially occurred through modulation of phospholipid synthesis and sphingolipid metabolism. These metabolomic profiling results provide new insights into the anticancer mechanisms of *E. indica* and facilitate the overall understanding of molecular events following therapeutic interventions.