Differentiation-inducing factor-1 inhibits growth and metastasis of triple negative breast cancer cells through AMPK-mediated inhibition of mTORC1-S6K signaling pathway

Fumi Tetsuo, Masaki Arioka, Koichi Miura, Sasaguri Toshiyuki

Kyushu Univ. Dept. of Clinical Pharmacol.

Differentiation-inducing factor-1 (DIF-1) identified in *Dictyostelium discoideum* has been reported to inhibit proliferation and motility of several cancer cells. However, the precise mechanism of its action remains undetermined. Here, we investigated whether DIF-1 could inhibit growth and metastasis of triple negative breast cancer (TNBC), the most aggressive and refractory phenotype of breast cancer, along with the underlying mechanisms involved. First, we performed experiments using cancer model mice made by injecting 4T1-Luc cells into mammary fat pad. Oral administration of DIF-1 significantly suppressed the primary tumor growth and spontaneous lung metastasis without adverse effects. In cultured 4T1-Luc cells, DIF-1 inhibited cell proliferation by suppressing STAT3-mediated cyclin D1 expression, and inhibited cell motility by suppressing the expression of Snail. DIF-1 did not reduce STAT3 mRNA or promote STAT3 protein degradation, suggesting that DIF-1 inhibited STAT3 protein synthesis. We revealed that DIF-1 inhibited the phosphorylation of S6K. Inhibition of S6K, using an mTOR inhibitors, also reduced the expressions of STAT3 and cyclin D1. DIF-1 rapidly and strongly phosphorylated AMP-activated protein kinase (AMPK) and its substrate Raptor, a component of mammalian target of rapamycin complex 1 (mTORC1). These results indicated that DIF-1 suppresses STAT3-cyclin D1-mediated proliferation and Snail-mediated motility of TNBC 4T1-Luc cells through AMPK-mediated inhibition of mTORC1-S6K signaling pathway.