CARP-1 FUNCTIONAL MIMETIC-4 IS A NOVEL SUPPRESSOR OF COLORECTAL CANCER CELL GROWTH AND METASTASIS

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ABSTRACT

Colorectal cancer (CRC) constitutes one of the most aggressive malignancies worldwide and in Malaysia. Due to high recurrence rate and toxic side effects of radiation and chemotherapies, new agents are urgently needed. CARP-1 is a peri-nuclear phospho-protein which plays a dynamic role in regulating cell growth and apoptosis. CARP-1 functional mimetics (CFMs) are a class of compounds that stimulate CARP-1. CFM-4, a lead compound, was shown to suppress growth and metastasis of various cancers. This study aimed to assess the extent to which CFM-4 inhibits CRC proliferation and metastasis and delineate the mechanism. CFM-4 anti-cancer effects of on CRC cells were investigated using MTT assay, Annexin V/Propidium iodide (PI) apoptosis assay, cell cycle analysis, quantitative real-time PCR (qRT-PCR) and Western blotting. Antimetastatic activities were assessed by migration, colony formation and invasion assays. CFM-4 inhibited CRC cell proliferation and was much more potent than the classical anti-CRC 5-fluorouracil. These effects were shown to be mediated at least in part by stimulating apoptosis, as indicated in our Annexin V/PI assay results. Cell cycle analysis showed that CFM-4 induced G2/M phase arrest. Molecularly, qRT-PCR results revealed that CFM-4 promoted intrinsic apoptosis by upregulating expression of *caspase-8* and *-9*, *p53, PUMA* and *Noxa,* and stimulated extrinsic apoptosis by enhancing the expression of death receptors. CFM-4 upregulated *NF-□B* signalling inhibitor A20-binding inhibitor protein-1 and the *PI3K* negative regulator *PTEN*. Western blot analysis results revealed that CFM-4 enhanced expression of CARP-1, caspase-8 and executioner caspase-3. Metastatic properties of the CRC cells were reduced by CFM-4 through blocking their capabilities to form colonies, migrate and invade through the matrix-coated membranes. The potent antitumor and anti-metastatic properties of CFM-4 against CRC are due to collective pro-apoptotic, anti-proliferative and anti-metastatic activities. Together our data warrants further investigations of CFM-4 as a potential anti-tumour agent for CRC malignancy and metastasis.

Keywords: Colorectal cancer; CARP-1; CFM-4; apoptosis; metastasis.

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