Docosahexaenoic acid modulates cell cycle and reduces cell proliferation by interference on WNT/Beta-catenin pathway in irradiated colorectal cancer cells

Taíssa Maria de Sá Ramos (API)[1], José Andrés Morgado-Díaz[1], Leonardo Borges Murad[1]

Grupo de Estrutura e Dinâmica Celular, Programa de Oncobiologia Celular e Molecular, Centro de Pesquisas – INCA, Rio de Janeiro, Brasil.

Email: leonardo.murad@inca.gov.br

ABSTRACTS

Colorectal cancer has been described as one of the main causes of death in the world, occupying the fourth type of cancer with higher mortality. Radiotherapy protocol in colorectal cancer demonstrated a high degree of effectiveness. However, radioresistance acquired by surviving cancer cells may lead to an adaptative state and promoting cell death evasion. Additionally, studies have pointed to beta-catenin, a fundamental protein in the activation of the canonical Wnt pathway, and its accumulation in the nucleus, as one of the main events responsible for radioresistance and tumor progression in colorectal cancer. Furthermore, recent studies have highlighted the role of DHA, a membrane lipid, as a regulatory agent of beta-catenin activity in some types of epithelial cancers. Thus, the objective of this work is to evaluate the effects of DHA associated with ionizing radiation on the localization of beta-catenin and its influence on tumor progression.

RESULTS

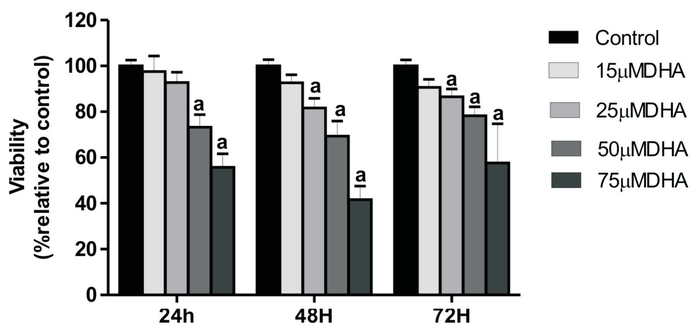

Fig.1. **Docosahexaenoic acid reduces cell viability in HT-29**
MTT assay demonstrated decrease on viability in colorectal cancer cell. HT-29 presented significant reduction in cell viability at 25μM, 50μM and 75μM concentration of DHA.

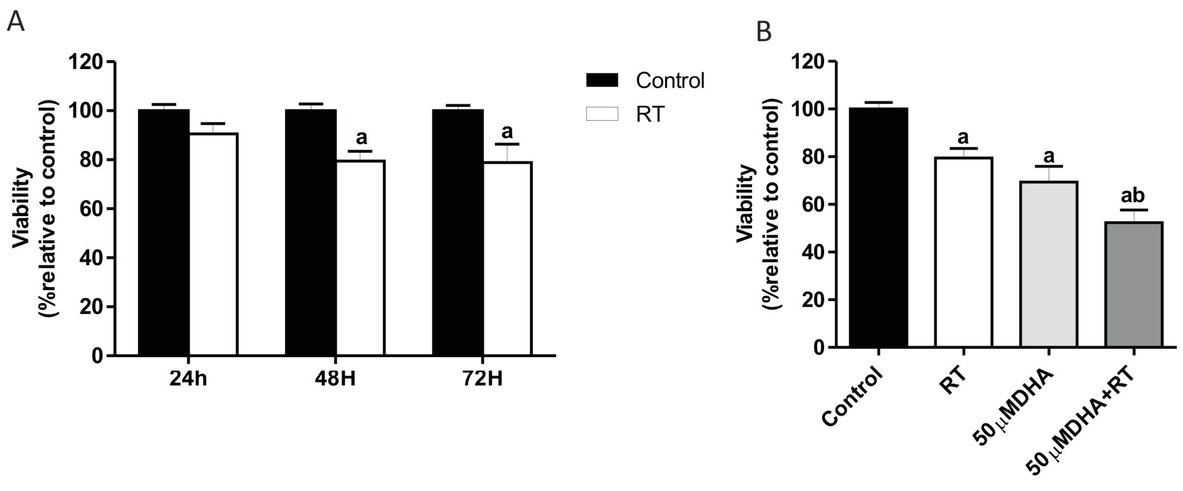

Fig. 2. **DHA potentiates irradiation effects in HT-29 cells**
(A) MTT assay showed decrease cell viability in HT-29 cell when exposed to radiation (5 Gy). (B) Cell viability significantly decreases in HT-29 treated with DHA + RT.

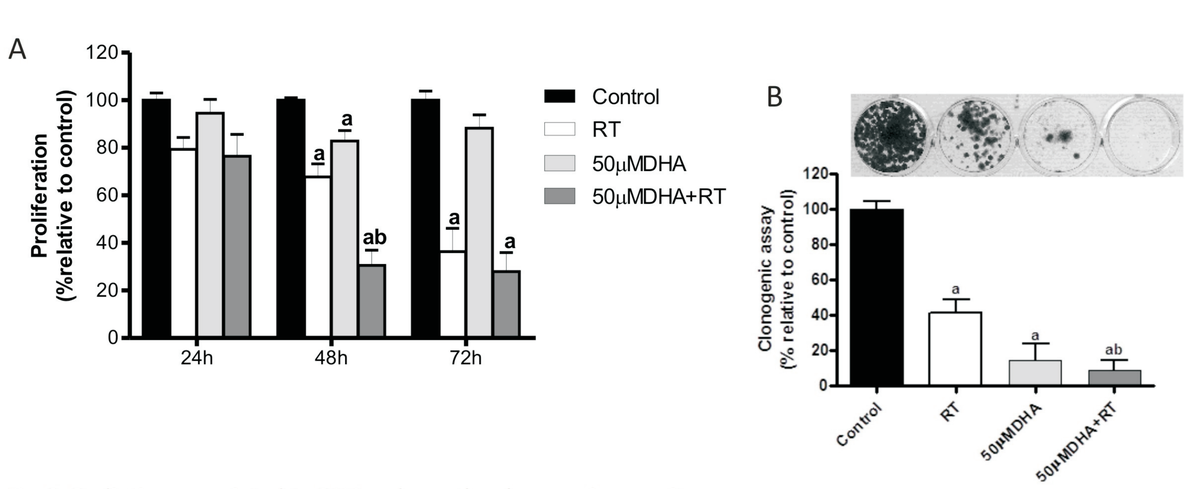

Fig. 3. **Radiation associated to DHA reduces the clonogenic capacity.**

(A) Proliferation capacity was potentially decreased in irradiated cells treated with 50M DHA. (B) Clonogenic assay demonstrated DHA capacity of inhibit colony formation after 11 days cell culture. Cells treated with radiation (RT) and 50M DHA presented smaller capacity of colony formation.

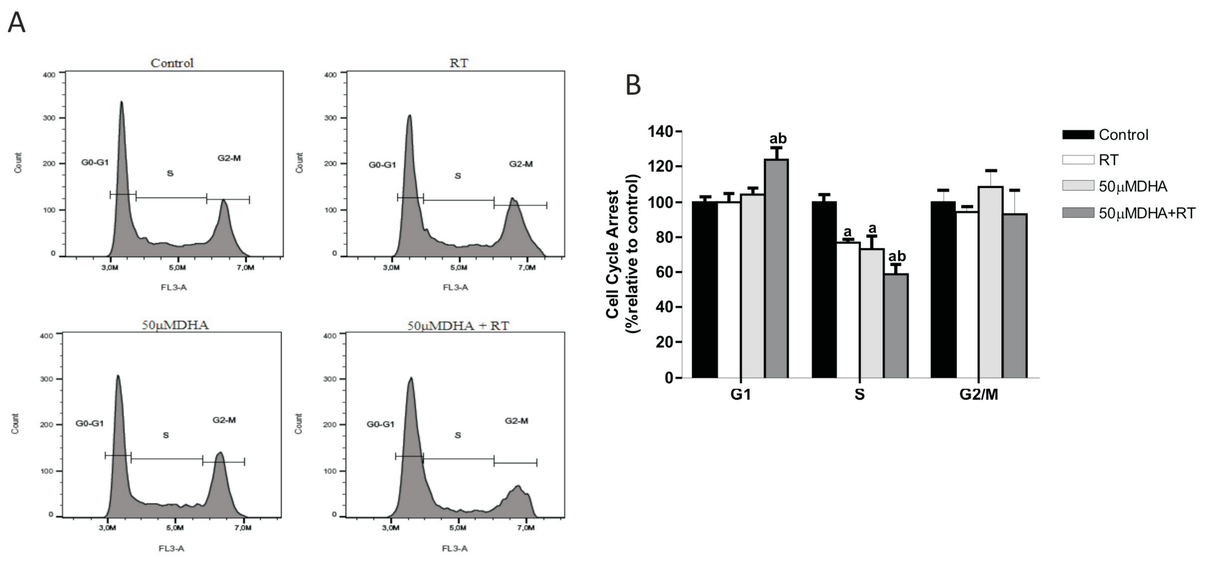

Fig.4. **Docosahexaenoic acid associated to radiation promotes G0/G1 phase cell arrest** (A) Graphical representation of citometry analysis. (B) Quantification of cell cycle analysis. Citometry analysis showed cell arrest in the G0/G1 phase and accumulation reduced in the S phase.

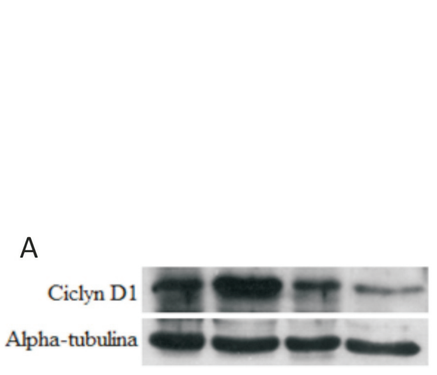

Fig.5. **Docosahexaenoic acid reduces expression of cyclin D1 in HT-29 cells**
(A) Western blot showed an alteration in cyclin D1 expression. (B) Statistical analysis revealed significant cyclin D1 increased in cells treated with RT alone. On the other hand, DHA+RT treatment reduced considerably the cyclin D1 expression.

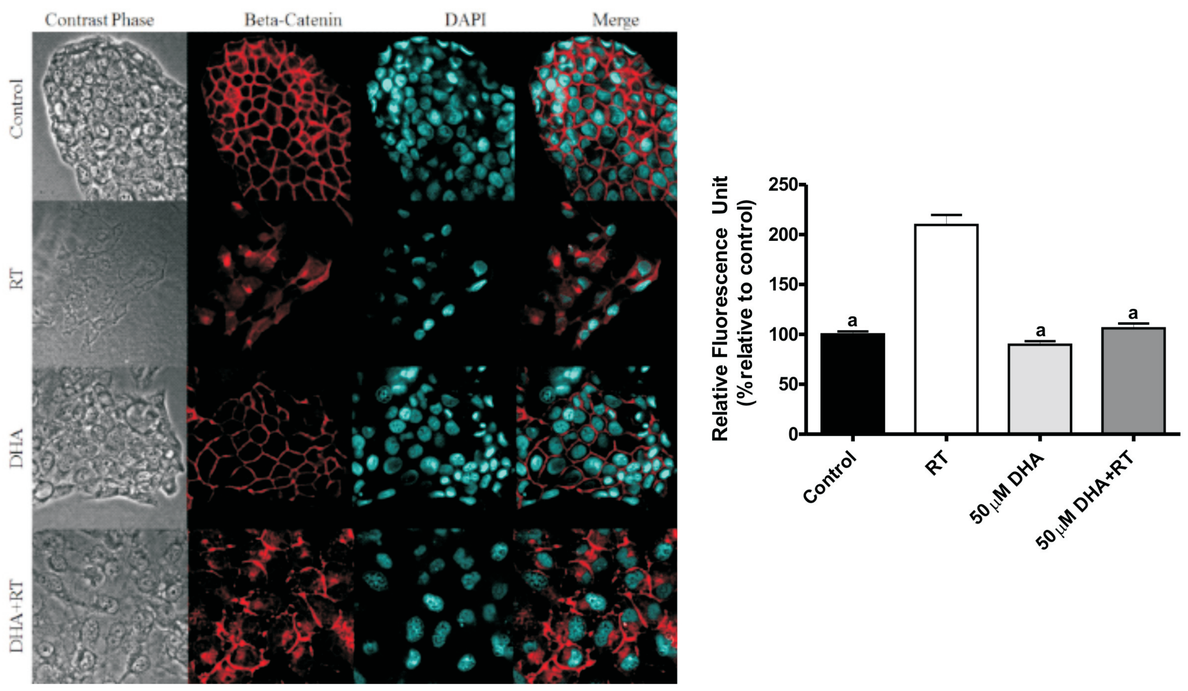

Fig.6. **Docosahexaenoic acid can inhibit beta-catenin nuclear translocation.** (A) Immunofluorescence indicated presence of beta-catenin (AlexaFluor488/Red color) in nucleus (DAPI/blue color) of the RT cells. (B) Signaling intensity analysis demonstrated significant reduction of beta-catenin in DHA+RT nucleus cells.

Projeto Gráfico: Serviço de Edição e Informação Técnico-Científica / INCA