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The Presence of GC-C in Extracellular Vesicles Secreted by Colorectal Cancer Cells

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Background: Guanylyl Cyclase C (GC-C) is a membrane-bound protein found on intestinal epithelial cells involved in the activation of CFTR. This protein has previously been involved in the development of colorectal cancer.

Extracellular vesicles (EVs) are bilayered vesicles of varying size (30 to 1,000 + nm in diameter) that believed to be secreted by all cells in the human body. In the past decade, EVs have garnered attention due to their impact in the field of oncology, where they have been shown to potentially serve as biomarkers for various cancers.

In this study, we looked at the EVs secreted by GC-C^+ and GC-C^- cell lines. We expected GC-C to be present on the EVs secreted by GC-C+ cell lines and that this finding may intake a role for GC-C at tissues distal to the intestinal epithelial cells.

Methods: GC-C^+ cells lines (T84 and CT26-hGCC) and GC-C^- cell lines (SW480 and CT26-WT) were cultured and their media was harvested, then ultracentrifuged to extract the EVs from the media. These EVs were then checked for the presence and absence of various markers (GC-C, Calnexin, TSG101) via Western Blot. Exosome size was assessed via NTA to further provide evidence for the identity of these EVs.

Results: Western blot confirmed the presence of TSG101 in both EV types samples, as well as the presence of GC-C in EVs derived from GC-C^+ cell lines, but not from GC-C^- cell lines. Calnexin was found to be absent in EV samples, excluding the possibility of lysate contamination. NTA analysis confirmed the correct size for the exosomes in sample.

Discussion: This study assessed the contents of EVs secreted by colorectal cancer cell lines. Our findings indicate the presence of GC-C on exosomes and microvesicles. Further studies will need to be conducted in order to assess the function of these GC-C^+ EVs in the setting of colorectal cancer.