

***Acinetobacter calcoaceticus* Shapes Epithelial-Macrophage Signalling in Colorectal Cancer**

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Background: The gut microbiota plays a critical role in colorectal cancer (CRC), yet the functional contributions of many bacterial species enriched within colorectal tumors remain poorly understood. Recent studies have identified *Acinetobacter* species in CRC-associated microbial communities; however, their potential role in promoting tumor-associated inflammation has not been defined. **Methods & Results:** To investigate the effects of *Acinetobacter calcoaceticus* on the colonic epithelium, murine CMT-93 colonic epithelial cells were treated with cell-free *A. calcoaceticus* supernatants. Exposure significantly increased the expression of the pro-inflammatory mediators MCP-1, IL-8, and TNF, which recruit and activate immune cells, like macrophages. To determine whether *A. calcoaceticus* also impacts macrophage, primary bone marrow-derived macrophages (BMDMs) were exposed to *A. calcoaceticus* cell-free supernatants. Treatment significantly increased IL-6 production, a pro-inflammatory cytokine implicated in epithelial proliferation and colorectal tumorigenesis. Fractionation of the supernatant revealed that the >10 kDa fraction of *A. calcoaceticus* secreted compound elicited increased IL-6 production in BMDMs, suggesting that the macrophage-stimulatory activity is associated with a higher-molecular-weight compound. We next investigated whether the macrophage response could influence the colonic epithelium. Conditioned medium collected from *A. calcoaceticus*-stimulated BMDMs was applied to CMT-93 cells and resulted in increased expression of the proliferation marker *Ki67*, supporting a potential macrophage-mediated effect on epithelial proliferation. To explore the clinical relevance of these findings, we analysed the Bacteria in Cancer database. CRC tumors enriched with *Acinetobacter* exhibited elevated expression of several proliferation and cancer-associated genes. Finally, we examined the inflammatory effects of *A. calcoaceticus* *in vivo*. Antibiotic-treated mice were orally gavaged with either PBS (vehicle control) or *A. calcoaceticus*. Colonized mice exhibited increased immune cell infiltration and elevated expression of pro-inflammatory cytokines in the colon. Furthermore, in mice treated with 3% DSS to induce colitis, *A. calcoaceticus* colonization significantly exacerbated DSS-induced intestinal pathology. **Conclusion:** These findings suggest that *A. calcoaceticus* drives inflammatory and proliferative signals that could participate in CRC development.