

## **The Interaction of *Clostridium perfringens* with Host Immune Cells**

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(ABSTRACT)

*Clostridium perfringens* is the most common cause of gas gangrene (clostridial myonecrosis), a disease that begins when ischemic tissues become contaminated with *C. perfringens*. *C. perfringens* quickly multiplies in ischemic tissues and spreads to healthy areas, leading to high levels of morbidity and mortality. As a species, the bacterium can synthesize thirteen different toxins. The alpha toxin (PLC) and perfringolysin O (PFO) are thought to be important virulence factors in gangrene. We wished to understand how *C. perfringens* is capable of avoiding killing by the host immune system, and determine if PLC and PFO play a role in this avoidance. We found *C. perfringens* was not killed by J774-33 cells or mouse peritoneal macrophages under aerobic or anaerobic conditions. Using electron microscopy, we showed that *C. perfringens* could escape the phagosome of J774-33 and mouse peritoneal macrophages. We believe the ability of *C. perfringens* to survive in the presence of macrophages is due to its ability to escape the phagosome. Using a variety of inhibitors of specific receptors, we identified those used by J774-33 cells to phagocytose *C. perfringens*. The scavenger receptor, mannose receptor(s), and complement receptor (CR3) were involved in the phagocytosis of *C. perfringens*. To determine if PFO or PLC were involved in the ability of *C. perfringens* to survive in the presence of macrophages, we constructed *C. perfringens* strains lacking these toxins.

The ability of *C. perfringens* to survive in the presence of J774-33 cells is dependent on PFO, while survival in mouse peritoneal macrophages is dependent on PFO and PLC. The ability of *C. perfringens* to escape the phagosome of J774-33 cells and mouse peritoneal macrophages is mediated by either PFO or PLC. Using a mouse model, we found that PFO and PLC were necessary for *C. perfringens* to survive *in vivo* using infectious doses 1000 times lower than those required to initiate a gangrene infection. We propose that PFO and PLC play a critical role in the survival of *C. perfringens* during the early stages of gangrene infections, when phagocytic cells are present and bacterial numbers are low.