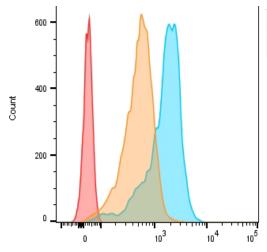


Oxidative Burst Analysis by Flow Cytometry

The growing number of monoclonal antibody therapies have provided better, more targeted treatment for the different types of cancers. With these exciting new therapies, the side effects of immune suppression have been declining over the years. However, even these new therapies can be associated with increased opportunistic infections [1-3]. The complex network of pathways and cells that encompass immune response regulation make the assessments of immune suppression one of the most challenging aspects to monitor in recovering patients. Understanding the mechanisms by which new therapies contribute to immune suppression and increase the incidence of opportunistic infections is a key aspect for immune-oncology.

Although there are no encompassing assays for the assessment of an immunocompromised state, there are assays to assess one of the key aspects of host defense. The Neutrophil and Monocyte Oxidative Burst Assay is a novel fluorometric analysis of host immune response to pathogens. As a normal immune response, macrophages survey their environment and through phagocytosis eliminate different pathogens. Once the pathogen is engulfed in a phagosome, a "burst" of superoxide species will flood the phagosome and eliminate the pathogen.

In order to assess such immune response *ex-vivo*, there has been a development of a small cell-permeable, non-fluorescent dye which transforms into a fluorescent compound as it encounters superoxide species. Such fluorescence can be observed through a plate reader, or flow cytometry as demonstrated in *Figure 1*.





Whole Blood treated with heat-killed Lysteria Monocytogenes (HKLM), PMA, or no treatment (NS), analyzed by flow cytometry.

This assay provides a simple yet powerful tool to study one key mechanism of immune defense associated with opportunistic infections. At CirQuest Labs, we offer the Oxidative Burst Assay that can be customized based on your needs.

Figure 1 Peak shift in Neutrophil Oxidative Burst Assay

- 1. Cheson, B.D., *Monoclonal antibody therapy of chronic lymphocytic leukemia*. Cancer Immunol Immunother, 2006. **55**(2): p. 188-96.
- 2. Focosi, D., et al., *Immunosuppressive monoclonal antibodies: current and next generation.* Clin Microbiol Infect, 2011. **17**(12): p. 1759-68.
- 3. Rafailidis, P.I., et al., *Infectious complications of monoclonal antibodies used in cancer therapy: a systematic review of the evidence from randomized controlled trials.* Cancer, 2007. **109**(11): p. 2182-9.