MULTIPLEX ASSAYS TO MEASURE INFLAMMATION MARKERS

Inflammation is a fundamental cellular process that is an immune response to pathogenic and non-pathogenic stimuli, and is a hallmark of various diseases including cancer, cardiovascular disease, diabetes, obesity and autoimmune disorders[1]. Monitoring the development and progression of inflammation has been shown to correlate with disease progression, so quantitative assays that measure levels of specific inflammation markers are widely used in preclinical and clinical studies[1]. The most common analytes are cytokines and interferons such as IL-1β, IFNγ and TNF-α. Additional markers are proteins that are produced during the acute phase of inflammation such as the C-reactive protein, which is a well-known liver inflammation marker[1].

Plate-based assays such as ELISA are quantitative, sensitive, rapid and cost-effective to measure levels of inflammation markers such as chemokines and cytokines. The method uses a sandwich format where capture antibodies coated on a plate capture the analyte of interest that is then detected using a second antibody and colorimetric, fluorescent or chemiluminescent readout. One of the most widely used assays is the MSD (Meso Scale Discovery) platform that uses a sandwich format and a unique electrochemiluminescent readout that is highly sensitive and specific[2]. Additionally, the MSD assay can be multiplexed so that multiple analytes can be measured from a single well – standard off the shelf assays can measure up to 10 analytes per well but the assay can be customized to include up to 40 analytes per well[3].

The MSD platform can support a diverse array of samples including cell culture media, biofluids such as serum from animal models and human patients. The assay can be used for various preclinical programs including PK/PD, toxicology and drug pharmacology studies performed in mouse, rat or nonhuman primate models. One of the main advantages of using the MSD assay is that it is highly sensitive and requires small sample volumes. This is important for samples from mouse or rat models where blood draw volumes are in the microliter range. Additionally, lower sample volumes are preferred for primates and other large animal models to simplify the blood draw process and minimize stress.

Due to the robustness of the MSD assay, there is significant interest in using the platform for large clinical and epidemiological studies. Studies have been published to demonstrate the reliability and robustness of the MSD assay and other platforms such as the Luminex bead-based assay. One study from 2017 tracked levels of 15 cytokines and chemokines in 250 people with HIV infection over 15 years[4]. The results showed that the MSD assay was able to detect most of the inflammatory biomarkers in about 80% of the samples which is considered to be acceptable and interestingly, the intra-individual variation suggested that the assay could be used to study different patient segments[4]. Another study evaluated a broad group of about 200 people that included men and women with diverse backgrounds to understand if the MSD assay was a reliable method to measure levels of circulating chemokines and cytokines[5]. The study measured 10 cytokines over a 4-month period and the data showed that specific biomarkers such as IL-6, IL-8, TNF-α, IL-10, IL-13, and IFN-γ could be reliably measured over the time period of the study[5].

In summary, it is clear that the MSD assay can be used for diverse applications ranging from early discovery, preclinical pharmacology, ADME and toxicology studies to clinical and epidemiological studies. The utility of the assay will continue to grow as additional off the shelf and custom panels are developed and validated for commercial use.

References:
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