Singulex®

GM-CSF (Granulocyte Macrophage Colony Stimulating Factor)

The Singulex GM-CSF assay greatly advances the clinical utility of GM-CSF in treating immune disorders. The assay's capability to detect over a broad dynamic concentration range in plasma enables efficacy and dosing studies using GM-CSF as a potential therapeutic agent.

Biology and Diseases

Granulocyte Macrophage-Colony Stimulating Factor (GM-CSF) is a cytokine produced by T cells, B cells, macrophages, mast cells and fibroblasts in response to inflammation or infection. Its primary role is to stimulate granulocyte and monocyte production in the bone marrow. Once GM-CSF has bound to its receptor, the activated complex propagates a signaling cascade that increases macrophage counts in order to fight infection. Thus increased GM-CSF can be observed during episodes of infectious disease and in a spectrum of inflammatory disorders. Conversely, a lack of GM-CSF resulting from autoantibody production has been linked to pulmonary alveolar proteinosis.

Therapies

Therapeutic use of GM-CSF has been investigated for pulmonary alveolar proteinosis, leukemia, neutropenia, breast cancer, hepatoma and Chron's disease. A recombinant form of GM-CSF (sargramostim) is marketed under the trade name Leukine. The drug is prescribed to adult patients with acute myelogenous leukemia following chemotherapy treatments. GM-CSF is able to boost neutrophil recovery and decrease the risk of infection due to a weakened immune system. In clinical settings, GM-CSF has been shown to enhance myeloid growth in the case of bone marrow transplantation. Furthermore, a phase III clinical trial will soon be underway to examine the safety and efficacy of GM-CSF for treating melanoma.

Unmet Need

GM-CSF continues to prove its usefulness as a prognostic biomarker in cancer settings and has been shown to effectively relieve inflammation caused by an overactive immune system. However, establishing baseline concentrations from healthy subjects still remains an obstacle with currently available ELISA and multiplex assay technologies. Studies have suggested that existing assays are not able to statistically differentiate the concentrations of plasma GM-CSF between healthy and diseased individuals. The future of diagnostics requires a sensitive immunoassay capable of stratifying the transition from healthy to diseased states based on minute changes in GM-CSF concentrations.

Singulex Answer

The Singulex GM-CSF assay, optimized for use on the Erenna System, enables scientists to measure over a broad dynamic range of GM-CSF in plasma. The assay has an LLoQ of 0.14 pg/mL and a reading range of 0.04-1000 pg/mL, providing accurate quantification of GM-CSF from only 100µL of plasma.

This assay will allow investigators to:

1. Measure the efficacy and dosing of therapeutics designed to increase levels of GM-CSF, particularly in restoring neutrophil production in leukemic patients.
2. Measure decreased concentrations of GM-CSF in patients with pulmonary alveolar proteinosis, which are below levels found in healthy subjects.
3. Design more robust clinical and preclinical studies when GM-CSF concentration is used as a therapeutic endpoint.
4. Understand how GM-CSF concentrations change in patients as they transition from healthy to diseased states.

ERENNA TECHNOLOGY ACCESS PROGRAM.

Through the Erenna Technology Access Program (ETAP), Singulex offers an interactive, results-driven solution to biomarker challenges faced by the pharmaceutical industry during product development. Singulex assists the development programs of our ETAP collaborators by developing customer-driven assays and access to a menu of fully-validated assays. Participants in ETAP gain access to the Singulex Erenna Immunoassay System, our proven expertise developing high-value immunoassays and our world-class customer support. Together with Singulex, our ETAP collaborators are expanding the utility of protein biomarkers and using them as tools to measure disease progression, drug efficacy and toxicity.

TABLE 1: Analytical sensitivity of the Singulex GM-CSF assay.

Lower Limit of Detection (LoD)	0.04 pg/mL
Lower Limit of Quantification (LLoQ)	0.14 pg/mL
Reading Range	0.04-1000 pg/mL

Erenna® System

TABLE 2: GM-CSF assay low-end standard curve data.

[GM-CSF] pg/mL	Detected Events	Std Dev	CV
4.5	2148	104	5%
2.25	1164	52	4%
1.13	624	32	5%
0.56	367	42	12%
0.28	234	18	8%
0.14	140	8	6%
0.07	98	6	6%
0.00	80	10	12%

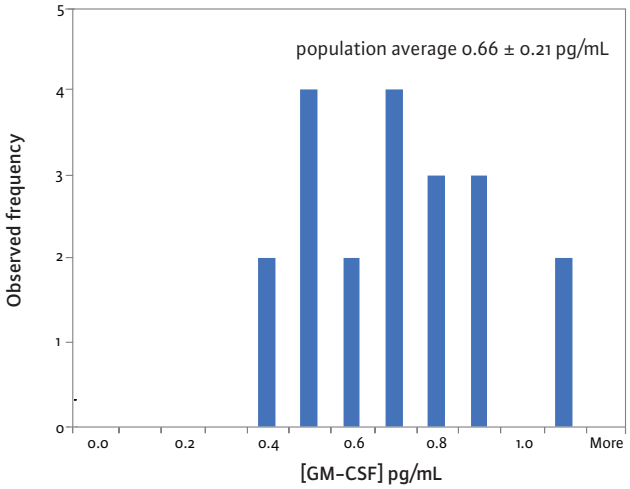

FIGURE 1: Plasma GM-CSF concentration in healthy human subjects.

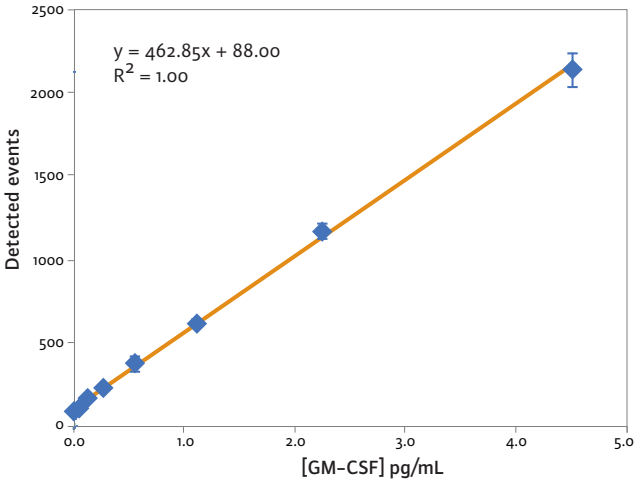

FIGURE 2: GM-CSF low-end standard curve signal.

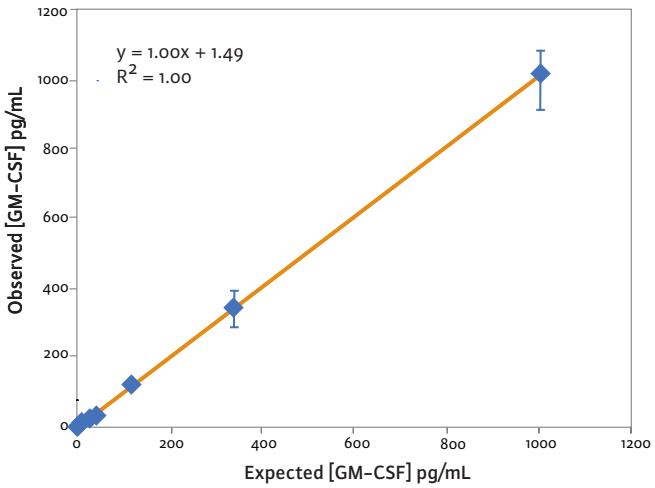

FIGURE 3: GM-CSF assay curve fit.

These standard curves are for representational purposes only. A standard curve must be run with each assay.

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