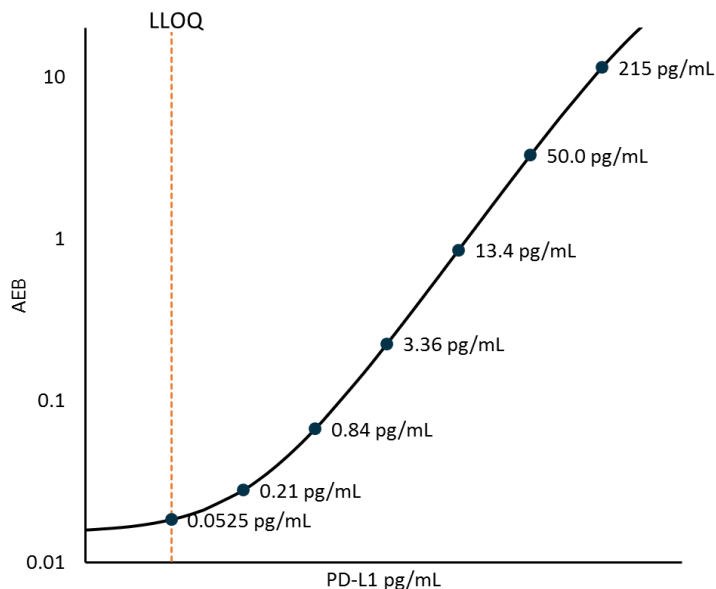


Description

PD-L1, or “programmed-death ligand 1” (also known as CD274 or B7-H1) is a membrane bound glycoprotein in the B7 family of cell surface ligands involved in regulation of the immune system. PD-L1 is expressed on a variety of inflammatory-activated cells, some carcinomas, and in melanoma (ovary, colon, lung, breast, and renal cell carcinomas). PD-L1 expression on tumor cells is correlated with poor prognosis in patients with cancers such as NSCLC, esophageal cancer, and pancreatic carcinoma. Levels of PD-L1 are increased in the plasma of cancer patients as well as in cerebrospinal fluid of gliomas. sPD-L1 is a biomarker of poor survival in patients with B cell lymphoma, renal cell carcinoma, metastatic melanoma or lung cancer, and is associated with advanced tumor stage. PD-L1 contributes to immune evasion by binding to PD-1 and CD80 to suppress the activation and proliferation of T cells and induce apoptosis of activated T cells. Blocking the PD-1/PD-L1 pathway to prevent this immune evasion and restore anti-tumor immunity has emerged as a promising anti-cancer strategy.

Calibration Curve: Calibrator concentrations and Lower Limit of Quantification depicted.



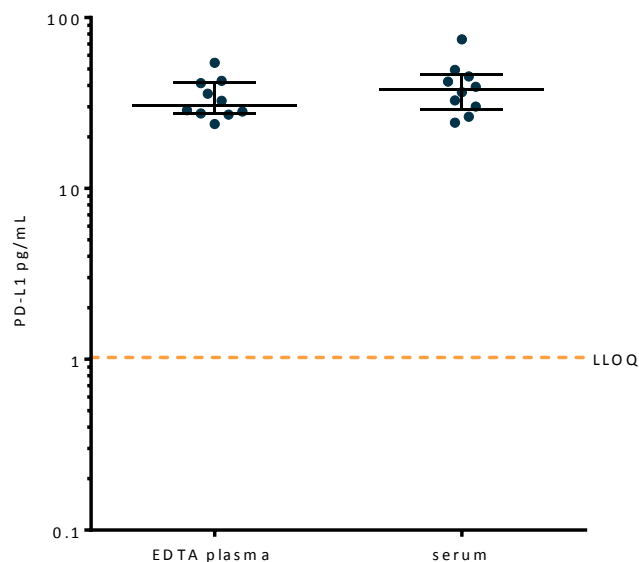
Lower Limit of Quantification (LLOQ): Triplicate measurements of serially diluted calibrator were read back on the calibration curve over 3 runs each for 1 reagent lot across 2 instruments (6 runs total).

Limit of Detection (LOD): Calculated as 2.5 standard deviations from the mean of background signal read back on each calibration curve over 3 runs each for 1 reagent lot across 2 instruments (6 runs total).

LLOQ	0.0525 pg/mL pooled CV 13% mean recovery 102%
LOD	0.0479 pg/mL range 0.0001-0.004 pg/mL
Dynamic range	0–4300 pg/mL
Diluted Sample volume (1:20 Dilution)*	100 µL per measurement
Tests per kit	96

*See Kit Instruction for details

Endogenous Sample Reading: Healthy donor matched EDTA plasma (n=10) and serum (n=10) samples were measured. Bars depict mean with SEM. Orange line represents functional LLOQ.



Sample Type	Mean PD-L1 pg/mL	Median PD-L1 pg/mL	% Above LOD
EDTA plasma	34.2	40.1	100%
Serum	30.6	38.0	100%

Precision: Measurements of 3 serum or plasma based panels and 2 calibrator based controls. Triplicate measurements were made for 3 runs each for 1 reagent lot across 2 instruments (6 runs total, 18 measurements).

Sample	Mean (pg/mL)	Within run CV	Between run CV	Between inst CV
Control 1	50.2	3.2%	6.6%	5.3%
Control 2	495	5.3%	5.9%	5.8%
Panel 1	43.3	3.8%	11.2%	9.6%
Panel 2	135	3.7%	7.8%	6.9%
Panel 3	44.2	4.6%	13.4%	0.1%

Note: Data in the following sections were obtained using the HD-1 Analyzer.

Spike and Recovery: 4 serum samples were spiked at high and low concentrations within the range of the assay and analyzed on HD-1.

Dilution Linearity: 1 spiked serum sample was diluted 2X serially from 20x (MRD) to 1280x with Sample Diluent.

Spike and Recovery	90% Range 72-98%
Spiked Dilution Linearity (1280x)	Mean = 106% Range: 99-112%

The Simoa PD-L1 assay kit is formulated for use on either the SR-X or HD-1 platform. Minor differences in performance claims between the HD-1 and SR-X may be observed when comparing datasheets for the two different platforms, due to experiments run at different time-points with different reagent lots and different samples. Data in this document was obtained from runs on the SR-X platform unless otherwise noted.