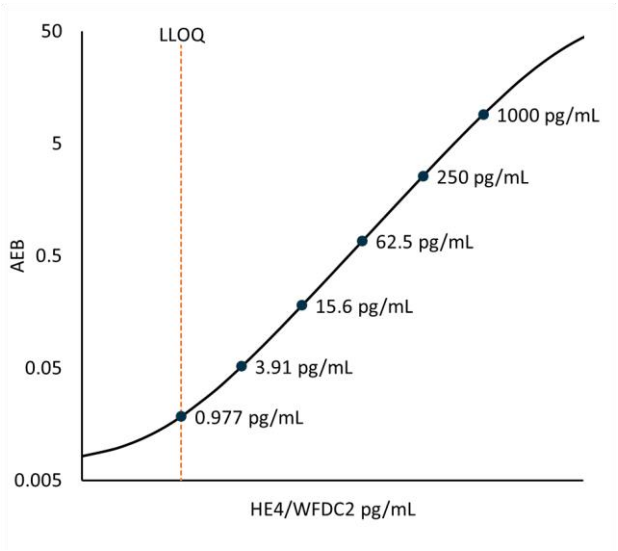
# Simoa® HE4/WFDC2 Discovery Kit
## HD-1/HD-X Data Sheet

Item 103059

### Description

Human epididymis protein 4 (HE4) is a 13 kDa protein coded by the gene *WFDC2*. Its mature glycosylated secretory form is approximately 25 kDa and consists of a single peptide and two whey acidic protein (WAP) domains containing a "four disulfide core" (FDC) encompassing eight cysteine residues. Although initially thought to have tissue-specific expression in the epididymis, HE4/WFDC2 is highly over-expressed in epithelial ovarian cancer (EOC) compared to normal ovarian epithelium. It has been recently demonstrated that over-expressed HE4/WFDC2 has a direct biological role in the promotion of OC cell proliferation, invasion and metastasis. HE4/WFDC2 levels have been monitored in EOC patients to determine recurrence or progression of the disease. HE4/WFDC2 has been used with an algorithm called Risk for Ovarian Malignancy Algorithm (ROMA) as a diagnostic tool to predict ovarian cancer. HE4 levels increase in other malignant neoplasms, especially of gynecologic (i.e., endometrial, tubal, vulvar cancer) and pulmonary origin. Accordingly, it has been proposed as a biomarker in other cancer types, in particular endometrial cancer and lung cancer. Many variables can affect the serum levels of HE4, such as age (higher in the elderly), smoking (higher by one-third in smokers than in non-smokers) and renal function.

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



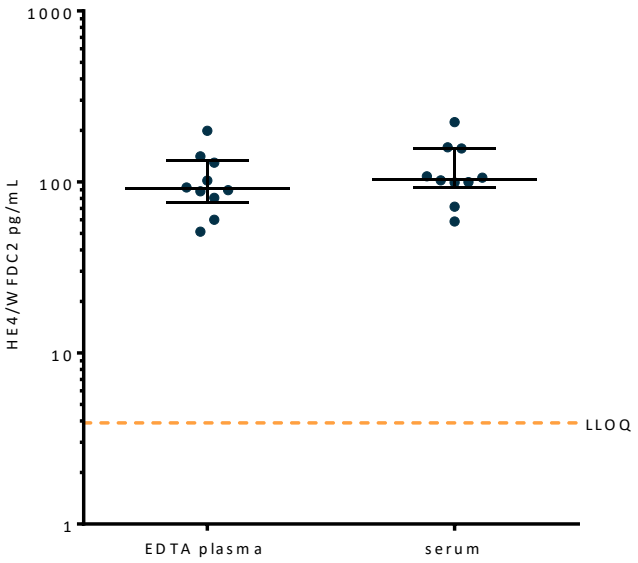

**Lower Limit of Quantification (LLOQ):** Triplicate measurements of serially diluted calibrator were read back on the calibration curve for 5 runs each for 1 reagent lot on a single instrument (5 runs total). The LLOQ is determined as the lowest dilution with a pooled CV ≤ 20% and sample concentration recovery between 80-120% of the expected.

**Limit of Detection (LOD):** Calculated as 2.5 standard deviations from the mean of background signal read back on each calibration over 5 runs for 1 reagent lot on a single instrument (5 runs total).

| | |
|---|---|
| **LLOQ** | **0.977 pg/mL**<br>pooled CV 10%,<br>mean recovery 98% |
| **LOD** | **0.135 pg/mL**<br>range 0.0281-0.274 pg/mL |
| **Dynamic range** | 0–4000 pg/mL |
| **Diluted Sample volume*** | 100 µL<br>per measurement |
| **Tests per kit** | 192 |

***See Kit Instruction for details**

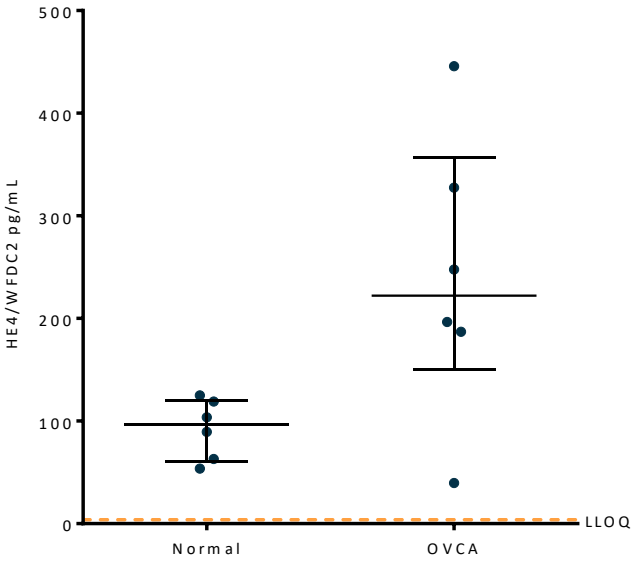
**Endogenous Sample Reading:** Healthy donor matched EDTA plasma (n=10) and serum (n=10) samples were measured. Bars depict median with interquartile range.




Quanterix Corporation
900 Middlesex Turnpike, Billerica 01821
techsupport@quanterix.com

| Matched human samples (n=10) | Mean HE4/WFDC2 pg/mL | Median HE4/WFDC2 pg/mL | % Above LOD |
|---|---|---|---|
| EDTA plasma | 104 | 91.2 | 100% |
| Serum | 119 | 104 | 100% |

**Normal vs Ovarian Cancer Sample Reading:** Healthy donor serum (n=6) and serum from patients with ovarian cancer (OVCA, n=6) were measured. Bars depict median with interquartile range.




**Precision:** Measurements of 3 serum or plasma based panels. Triplicate measurements were made for 5 runs using 1 reagent lot and a single instrument (5 runs total, 15 measurements).

| Sample | Mean (pg/mL) | Within run CV | Between run CV |
|---|---|---|---|
| Panel 1 | 177 | 2.7% | 1.9% |
| Panel 2 | 1160 | 3.5% | 3.3% |
| Panel 3 | 451 | 3.6% | 3.0% |

**Spike and Recovery:** 1 EDTA plasma sample and 3 serum samples were spiked at high and low concentrations within the range of the assay.

**Dilution Linearity:** 1 spiked EDTA plasma sample and 1 endogenous serum sample were diluted 2x serially from MRD (4x) to 256x with Sample Diluent.

| | |
|---|---|
| **Spike and Recovery (Serum/Plasma)** | **108%** Range 99-121% |
| **Spiked Plasma Dilution Linearity (256x)** | **Mean = 107%** Range: 105-109% |
| **Endogenous Serum Dilution Linearity (256x)** | **Mean = 106%** Range: 99-115% |

**Specificity:** Normal serum (n=2) samples were directly incubated with detector antibody and run at MRD. Average knock-down was **97%**.

Quanterix Corporation
900 Middlesex Turnpike, Billerica 01821
techsupport@quanterix.com