

# NovaSeq™ 6000 SP vs S4 flow cell comparison

## Background

The Olink® Explore platform enables protein biomarker analysis of your samples based on PEA technology coupled with NGS readout, using the high performance Illumina® NovaSeq™ 6000 Sequencing System.

When Olink® Explore was first launched, each sequencing run was performed according to a standard Illumina Sequencing Recipe in combination with the NovaSeq S1 flow cell with the Illumina control Library (PhiX) spiked into every Olink Library.

### Definition

A **library** is a pool of DNA fragments with attached DNA adapters that are designed to interact with a specific sequencing platform.

### Definition

The **flow cell** is where the sequencing chemistry occurs. It is a glass slide that contains nano-wells. Each nano-well contains oligonucleotides that provide an anchoring point for the DNA adapters to attach to.

In the technical note *NovaSeq™ 6000 SP vs S1 flow cell comparison* we showed that the SP flow cell gives the same quality of results as the S1 flow cell.

The flow cells SP and S1 have two lanes each. The S4 flow cell has four lanes. Therefore the complete Olink Explore 1536 library can be run on one S4 flow cell, while it would take two flow cells run in parallel using SP or S1.

To be able to offer a faster and more simplified sequencing run at a reduced cost and without compromising on data quality, Olink has improved the workflow by introducing a Custom Sequencing Recipe together with Illumina. The Custom Recipe enables sequencing of Olink Libraries without the addition of control Library (PhiX), using the NovaSeq S4 flow cell.

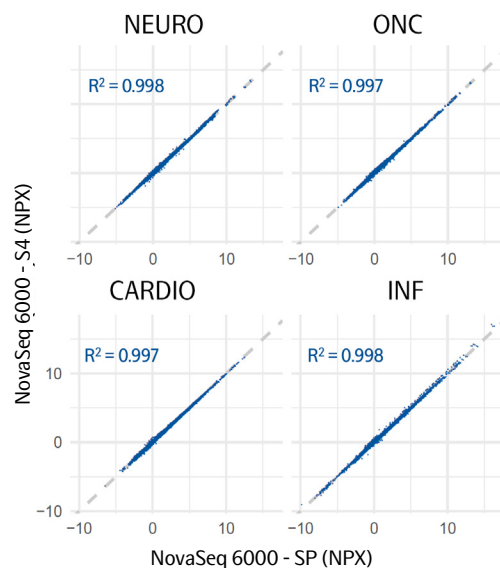
## Method

There were 88 samples used in the evaluation of the performance of Explore 1536 on the SP vs S4 flow cell. More than half of these were from healthy, normal subjects. Four Olink® Explore 384 Libraries were prepared, one for each of the four panels included in an Olink® Explore 1536 Reagent Kit (Explore 384 Inflammation, Explore 384 Cardiometabolic, Explore 384 Oncology and Explore 384 Neurology). The four Olink Explore Libraries were sequenced, using both the SP and S4 flow cells. Samples were kindly provided by Prof. Mathias Uhlén (Royal Institute of Technology, Sweden).

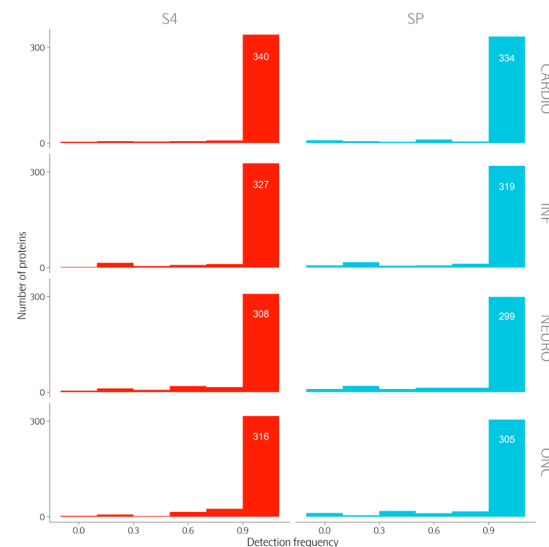
## Results

The NPX values between samples run using an SP versus an S4 flow cell were highly correlated as shown in Figure 1. There were no block or panel effects identified.

The detectability for each assay was compared between the two flow cells and similar results were attained, as shown in Figure 2.



**Figure 1** NPX results for a full plate of plasma samples run on Olink® Explore 1536 with an S4 flow cell versus an SP flow cell.



**Figure 2** The detection frequency for the assays on the four different Explore 384 panels (Cardiometabolic, Inflammation, Neurology and Oncology).

These results show that the SP flow cell gives similar quality of results as the S4 flow cell, and that the number of reads is sufficient when Olink Libraries are sequenced without the addition of PhiX.



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