Technical note

Concordance and repeatability tests to evaluate lab performance

Introduction

The purpose of the concordance validation was to evaluate if the same high quality of data could be obtained at Olink™-trained labs as at Olink Analysis Service labs, to ensure they can provide stable and accurate results that fulfill the Olink QC acceptance criteria.

Study design

The same set of samples was run on Olink™ Target 48, Olink™ Target 96 and Olink™ Explore panels at Sequanta Technologies in Shanghai, China, and at Olink Analysis Service in Boston, US.

Operator 1 from Sequanta ran both the Target and Explore panels, while Operator 2 ran the Target panels only, and is therefore noted as N/A in the Explore entries in the results tables.

The detectability, CV and correlation were compared between the two sites and the evaluation was performed by the Olink Data Science team.

Samples

The sample set consisted of samples from patients with various diseases, as well as healthy donors. See Figure 1 for more information about samples types.

Table 1. The sample distribution for the different Olink panels.

Olink™ panel	Number of samples	Subjects
Target 48 Cytokine	16 samples (14 plasma and 2 serum)	12 patients 4 healthy donors
Target 96 Neuro Exploratory	16 samples (14 plasma and 2 serum)	12 patients 4 healthy donors
Target 96 Oncology II	16 samples (14 plasma and 2 serum)	12 patients 4 healthy donors
Explore 384 Cardiometabolic	40 samples (36 plasma and 4 serum)	30 patients 10 healthy donors

Results

All runs passed the Olink QC criteria. Refer to the software user manuals for [Olink NPX Signature](#) and [Olink MyData Cloud](#) for detailed information about the different QC criteria.

Detectability

Detectability of each assay was calculated as the percentage of samples with results above the estimated limit of detection (LOD), or, in the case of Olink Target 48, above the lowest quantifiable level (LQL).

Table 2. The average detectability of each panel by lab and operator

Olink™ panel	Olink Analysis Service lab	Sequanta Operator 1	Sequanta Operator 2
Target 48 Cytokine	86%	87%	86%
Target 96 Neuro Exploratory	84%	80%	81%
Target 96 Oncology II	97%	99%	99%
Explore 384 Cardiometabolic	94%	97%	N/A

Intra CV comparison

The coefficient of variance (CV) was calculated on a per assay basis, using the control samples that were included in each run (see Table 3 and 4). For this calculation, data below LOD or LQL was excluded.

Table 3. The average CV of each panel by lab and operator.

Olink™ panel	Olink Analysis Service lab	Sequanta Operator 1	Sequanta Operator 2
Target 48 Cytokine	5.1%	3.2%	3.7%
Target 96 Neuro Exploratory	5.0%	6.6%	7.1%
Target 96 Oncology II	3.2%	3.1%	4.1%
Explore 384 Cardiometabolic	13.6%	7.5%	N/A

Table 4. Quantified values and CVs of IL18 for each sample run on Olink™ Target 48 in both labs. CVs are presented for all three values from both labs.

Sample	Olink™ Analysis Service (pg/mL)	Sequanta Operator 1 (pg/mL)	Sequanta Operator 2 (pg/mL)	CV
Healthy serum	242	247	251	1.7%
ALS	380	415	392	4.6%
Healthy serum	492	442	441	6.4%
Solid tumor cancer	183	207	198	6.2%
Healthy plasma	309	308	290	3.5%
Follicular lymphoma	597	594	566	2.9%
Healthy plasma	153	147	134	6.6%
Lung cancer	631	711	615	7.8%
CAD	209	225	206	4.8%
Breast cancer	427	553	538	13.6%
Migraines, Hypertension	869	931	875	3.8%
Type II Diabetic donor	372	371	354	2.9%
Hypertension, Atrial fibrillation	299	294	280	3.4%
Rheumatoid arthritis	377	390	373	2.4%
Alzheimers disease	344	419	351	11.2%
Gout	623	737	704	8.5%

Correlation

Correlations of NPX values between Analysis Service and Sequanta were calculated on a per assay basis for Olink Target 96 and Olink Explore. For Olink Target 48, the correleation of absolute concentrations in pg/mL was calculated. Each operator's correlations were calculated separately. For this calculation, data below LOD or LQL was excluded.

Table 5. The average correlation values (r, Pearson correlation) of each panel by lab and operator.

Olink™ panel	Olink Analysis Service & Sequanta Operator 1	Olink Analysis Service & Sequanta Operator 2
Target 48 Cytokine	0.96	0.97
Target 96 Neuro Exploratory	0.85	0.91
Target 96 Oncology II	0.92	0.92
Explore 384 Cardiometabolic	0.94	N/A

PCA plots

Scatter plots along the two principal components were generated to compare global data performance. Figure 1 displays PCA plots from both labs combined to evaluate sample clustering across labs. When looking at the PCA of results from both labs combined, the same samples tend to cluster with themselves suggesting that the labs are performing similarly.

Conclusion

The results showed that Sequanta could produce the same high quality data as the Olink Analysis Service lab.

The small variances seen can be explained by sample shipment and storage conditions as well as differences between kit lots.
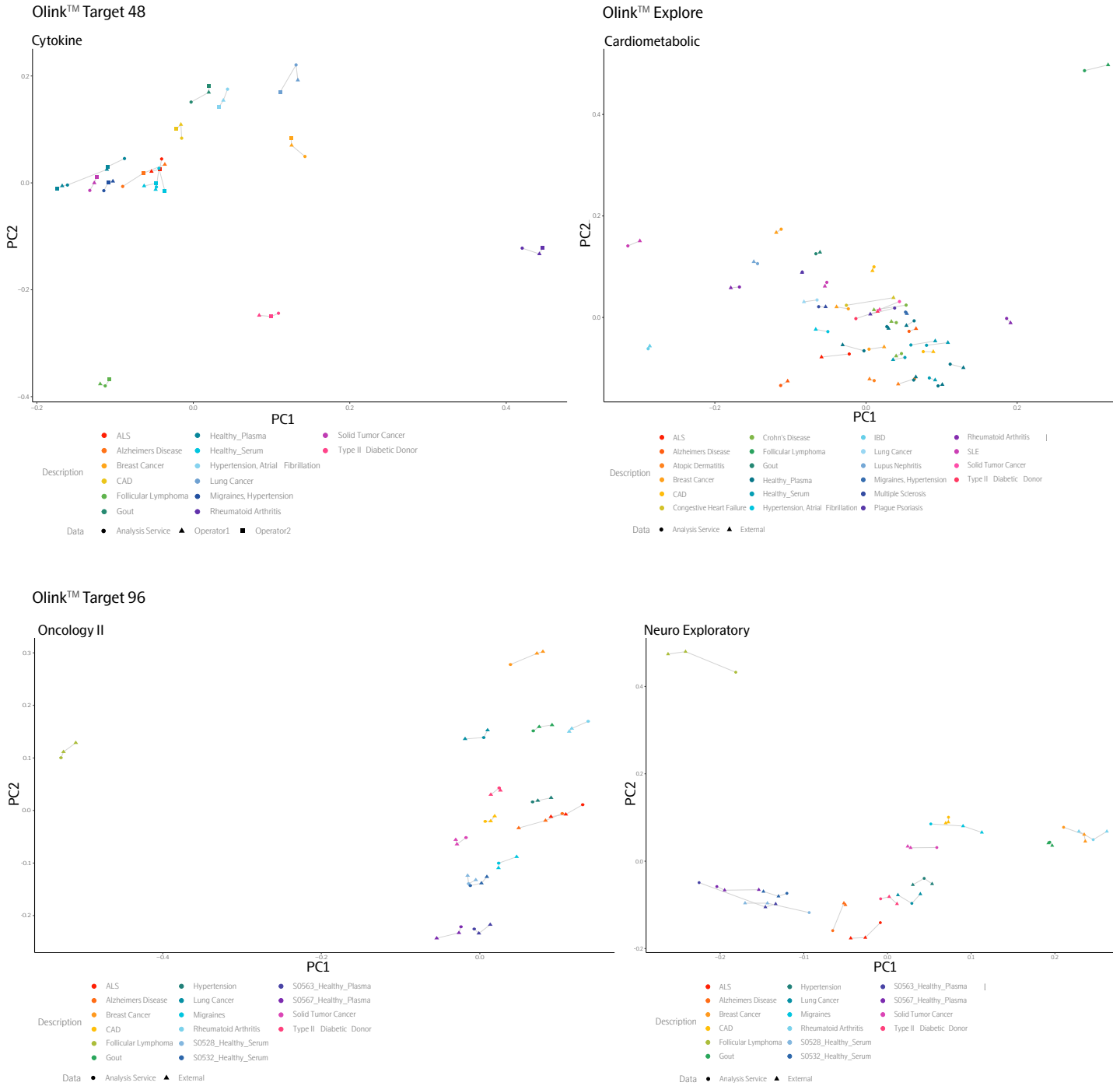
Figure 1. PCA plots combining the Olink™ Analysis Service and Sequanta lab results to evaluate sample clustering across labs for several Olink panels, colored by sample description and shape representing lab and operator. Lines connect the same sample replicates.

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