

PluriTest™ Report

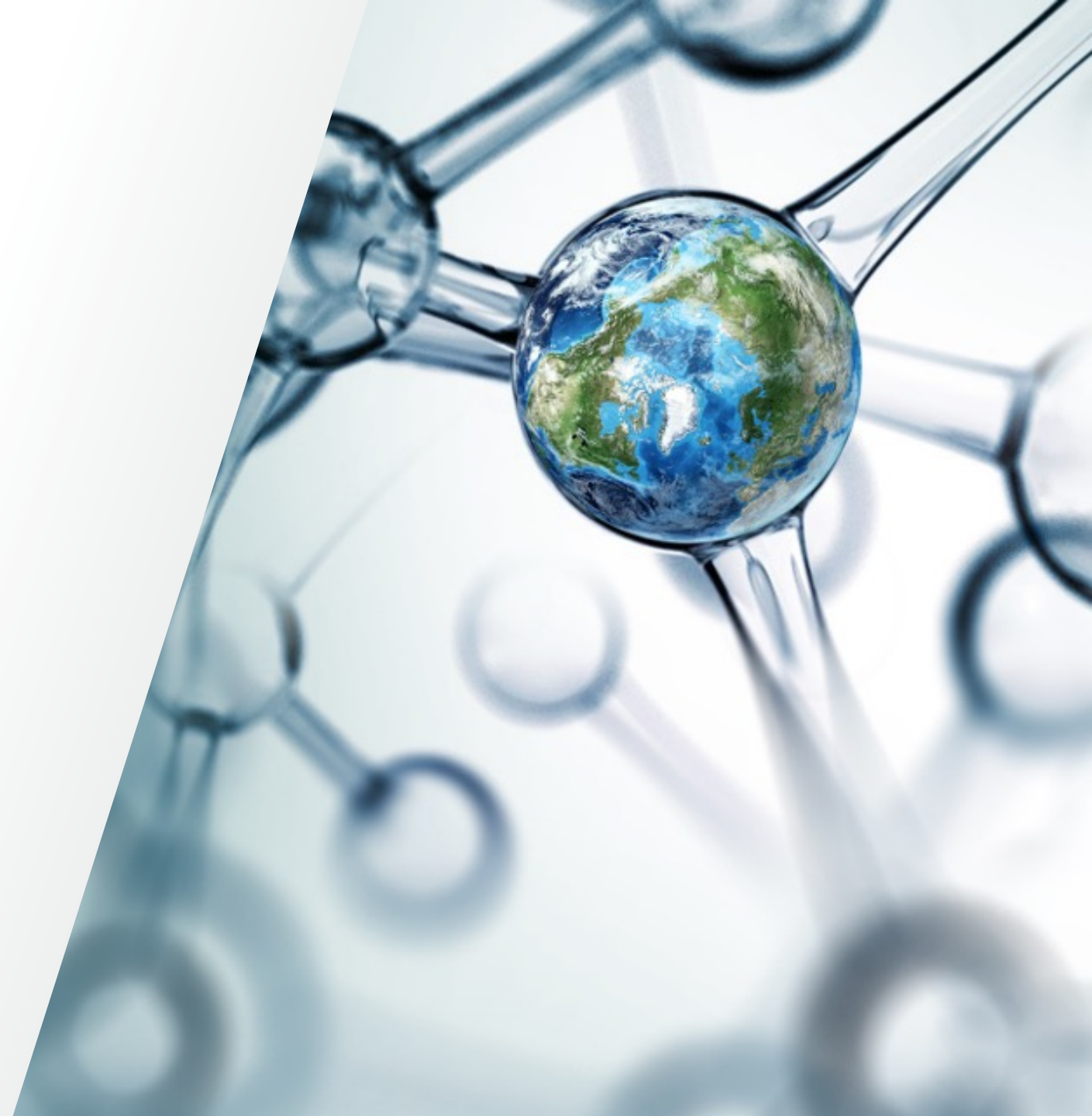
Client Name: Johns Hopkins Hospital

Quote No: 11758283 SO

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Summary of Services

Project Summary:

Johns Hopkins Hospital (Client) is interested in Services provided by the Life Technologies Corporation in the analysis of two (2) Client provided samples using the PluriTest™ Service. In this assay, 36,000 transcripts and variants against a >450 sample reference set are assessed for gene expression analysis.

Service Description:

The transcriptome of all samples were analyzed and processed in the PluriTest™ algorithm to generate a Pluripotency and Novelty Score

- The Pluripotency Score is based on many samples (pluripotent, somatic, and tissues) in the stem cell model matrix which consists of an extensive reference set of >450 cell/tissue types, including 223 hESC lines, 41 iPSC lines, somatic cells, and tissues. Samples with positive pluripotency values are more similar to the pluripotent samples in the model matrix than to all other classes of samples in the matrix.
- The Novelty Score is based on well-characterized PSCs in the stem cell model matrix. A low novelty score indicates that the tested sample can be well reconstructed based on existing data from other well-characterized iPSC and ESC lines. A high novelty score indicates that there are patterns in the tested sample that cannot be explained by the existing database of well-characterized, karyotypically normal pluripotent stem cells.
- Partially differentiated pluripotent cells, teratocarcinoma cells, or karyotypically abnormal embryonic stem cells may have a high pluripotency score but cannot be reconstructed well with data from well-characterized, normal pluripotent stem cells, and thus are expected to have a high novelty score.
- **Borderline Results:**
- Samples that are flagged as borderline rather than pluripotent is an indication that the tested sample(s) have a molecular signature that is slightly different from the database samples. The database samples are based on a limited number of PSC lines, so it is not entirely unlikely that a sample may differ. Since there are many different ways to reprogram cells, the core pluripotency may be maintained, but there could be molecular or epigenetic differences affecting a portion of the genes.

Materials & Methods:

1. RNA Purification:
 - Cells were prepared according to the PureLink™ RNA Mini Kit (Catalog #: 12183025) and quantified using the NanoDrop™
2. GeneChip® Preparation:
 - 100 ng total RNA was used to prepare the GeneChip® for the PluriTest™

PluriTest™ Results



- 1. Sample PT-5979 was found to be pluripotent.
- 2. The raw gene expression data for these samples will be delivered to the client.

Sample	Sample ID	PluriTest Result	PluriCor	NovelCor
PT-5979	CAVC-C11 Pluritest	Pass	38.9534	1.429537
PT-5981	iPSC	Pass	39.41394	1.209475
PT-5982	Non_iPSC	Fail	-43.40809	2.714002

Table 1: PluriTest™ results. The samples were analyzed using an algorithm that integrates gene expression data to authenticate pluripotency status. Samples are screened against samples in the stem cell database and given a pluripotency score (PluriCor) and novelty score (NovelCor), which are shown in the table. Pass shows a clear pluripotency signature. Fail means the samples are not pluripotent. A non-iPSC sample was used in this experiment to serve as a negative control for non-pluripotency.

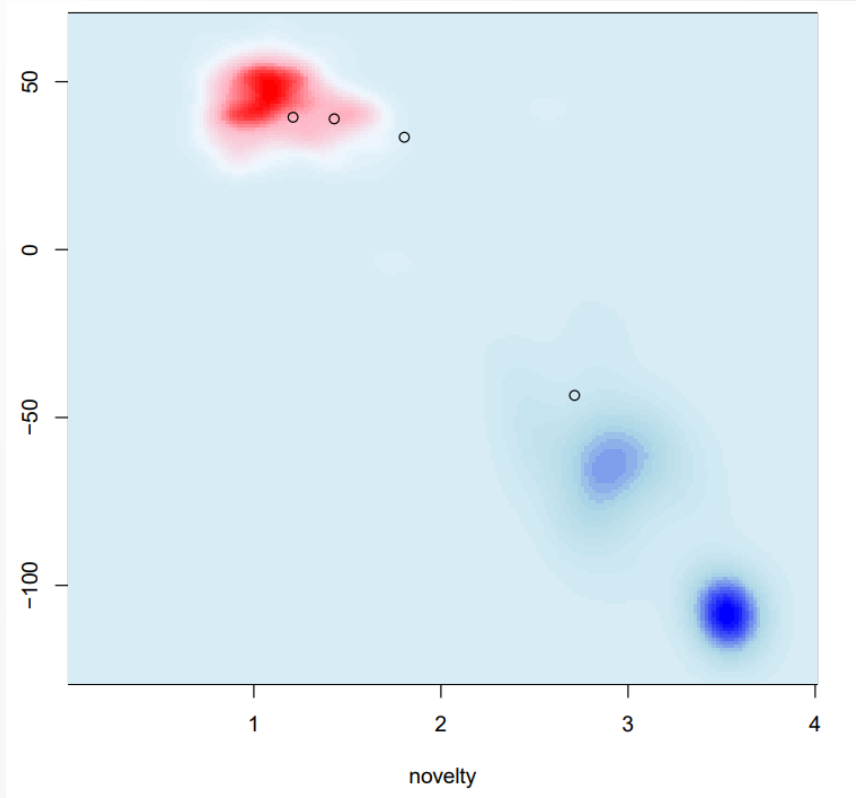


Figure 1: Pluripotency Plot. The pluripotency plot window provides a visual representation of the tested samples in the analysis. The pluripotency and novelty x/y scatter plot combines the pluripotency score on the y-axis with the novelty score on the x-axis. The red and blue background hint to the empirical distribution of the pluripotent (red) and non-pluripotent (blue) samples in the reference data set. The distribution of the samples (Table 1) can be seen in the above figure. A non-iPSC sample was included in this experiment to serve as a negative control representing non-pluripotency.