

From: [John West](#)
To: [Richard Chen](#)
Subject: Draft of what we might offer as Laboratory Assays
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Attachments: [Laboratory assays JW 13Jan2012.ppt](#)

Hi Rich,

I have put together an outline of the Laboratory Assays we might offer. I don't think we need this level of detail at the Board meeting, but we should discuss this internally, starting with your thoughts. In addition, some of this should be the subject of patent filings, so we should not circulate it outside the company until we do that.

John West

Laboratory Assays - Overview

- Generic genome-scale assays which can be applied to any sample without prior knowledge of the sample
 - Whole genome Plus
 - Exome Plus
 - Genotyping Plus
- Custom validation of key results from specific genomes
- “Virtual genomes” directly inferred from sequence data of others (e.g. children from parents)

Laboratory assays – Whole genome Plus

- Illumina or CGI whole genome sequence with short-insert paired-end reads (may be provided by customer). Coverage & read length recommendations TBD
- Targeted re-sequencing using array or oligo-based pullout:
 - Additional coverage to fill in systematic gaps due to bias of biochemistry / molecular biology used for whole genome
 - Long-fragment pullout to resolve repeats, particularly in approx 3,000 genes with alignment problems due to gene-family homologies
- Whole genome and/or exome-focused genotyping to provide:
 - Technologically orthogonal validation of numerous specific medically interpretable alleles
 - Calibration of the sequencing error model
 - Independent assessment of Copy Number Variation
- Electrophoretic assay of certain VNTR's which are difficult to assess with NGS
- FISH assay of chromosomal translocations, difficult to assess with NGS
- RNA-seq from same sample, for expression, allelic bias, genetic variant-driven splice variation, partial phasing of individual genomes

Laboratory assays – “Exome plus”

- Exon re-sequencing by pullout from customer sample (may be provided by customer)
- Additional re-sequencing by pullout, to capture regions outside the exons which are in VariMed, Regulome, lincRNA catalog & possibly others
- Other elements from the list of assays which supplement the Whole Genome product. Selection likely to be based on customer’s specific goals and pricing

Laboratory assays – Genotyping Plus

- Ability to run Personalis' interpretive content against a large set of genome-wide genotyping data sets a customer may already have, potentially to select samples for more in-depth (sequencing based) assessment
- Depending on the costs and the interest in this, we could also have a custom array made to capture more of the Personalis content

Laboratory assays – Validation of key results from specific genomes

- Sanger re-sequencing of variants which underlie key conclusions of a genome interpretation project.
 - Targeting of regions to re-sequence can be by PCR, or may require more complex methods in homologous regions.
 - For highest confidence, combine forward & reverse Sanger reads of each amplicon.
- Targeted Sanger re-sequencing of full-length cDNA to confirm phasing of compound heterozygous variant-pairs which underlie key conclusions of a genome interpretation project
 - Depending on spacing of the variant pair on a cDNA, this may need to be paired-end Sanger sequencing
- Other assays to confirm key variations not amenable to Sanger sequencing (e.g. VNTR's, certain CNV's, certain balanced translocations)

Virtual Genomes

- Genomes directly inferred from the data of others who have been sequenced (e.g. children from parents or vice versa)
- Inference based on recombination, mapped by genotyping or from the sequence data itself
- Can capture de novo mutation events
- Can provide genomes of individuals already deceased
- Whole genome data sets at a cost approaching exomes