OICR Genomics Accreditation and Services

Accreditation

Clinical trials are increasingly requiring participant labs to be accredited by one or more internationally recognized organizations. Accreditation is evidence that the lab has a fully developed Quality Management System, appropriately validated assays, qualified testing personnel, suitable facilities, controlled vendor relationships, and more. Typically, less-complex validated assays are the focus of accredited labs (e.g. detecting a single analyte in a blood sample), so offering Whole Genome and Transcriptome Sequencing makes us one of only a handful of international labs with this distinction. Genomics and Tissue Portal are accredited by the College of American Pathologists (CAP), which is the preferred organization for many US-based clinical trials, by Accreditation Canada Diagnostics (ACD), and by the Clinical Laboratory Improvement Amendments of 1988 (CLIA). ACD is a Canadian accreditation organization that audits directly against the ISO 15189 Medical Laboratories standard, and labs who wish to obtain a commercial lab license must first obtain this accreditation. Ultimately, the three accreditations are a mark of excellence that allow the lab to participate in major clinical trials, and it opens the door to eventually offer diagnostic services for reimbursement from OHIP, should OICR ever determine that this activity is appropriate.

ACD Additional Details: Accreditation Canada Diagnostics (ACD) accreditation symbolizes the lab's commitment to quality, continuous improvement, and scientific/business best practices. It acknowledges that we are compliant with the ISO 15189 Medical Laboratories standard, which is a set of requirements for appropriate operation of a medical diagnostic laboratory. ISO 15189 covers topics such as safety, assay validation and performance, pre/post-analytical techniques, training, informatics/IT, facilities, equipment, quality management, organizational structure, etc. Accreditation provides confidence to clients and collaborators that the lab offers a high-quality product, and many clinical trials are now requiring accreditation as a condition for participation. OICR Genomics is one of a very small number of international labs with a validated whole genome and transcriptome sequencing assay, which is a major achievement due to the complexity of (and demand for) such an assay.

Service Descriptions

WGTS

OICR Genomics offers a Whole Genome and Transcriptome Sequencing (WGTS) validated clinical assay, performed in our ISO-compliant laboratory, which is accredited by the College of American Pathologists (CAP), Accreditation Canada Diagnostics (ACD), and the Clinical Laboratory Improvement Amendments (CLIA). The resulting clinical report covers a subset of SNVs, SVs, fusions and CNVs implicated in cancer. The reported variants are derived from OncoKB, which is a precision oncology knowledge base containing information about the biological effect, prevalence and prognosis and treatment implications of specific cancer gene alterations. Raw data is also available to meet the customer's research needs.

The WGTS assay is also available for research use only (RUO), processed through the same processes and pipelines as the accredited assays, with the same data returned to the customer. Clinical reports are not provided for RUO customers.

Whole genome (WG) sequencing: DNA extraction from fresh frozen (FF) or formalin-fixed paraffin-embedded (FFPE) tissue, cells, and blood (buffy coat) is performed using the Qiagen Puregene Blood Kit and/or the Qiagen Allprep DNA/RNA FFPE Kit and quantitated using the Qubit 4.0 instrument. The KAPA Hyper Prep Kit (Roche) is used to create whole transcriptome libraries, which are then visualized and quantitated using the Fragment Analyzer and Qubit 4.0. Library quality is validated using shallow MiSeq

sequencing to assess mapping to genomic regions. Whole genome libraries are then sequenced using 2x150 paired reads on an Illumina NovaSeq6000 system to the requested target depth. Clinical whole genomes are sequenced to 30X, 40X or 80X depending on customer requirements and the purity of the tumour.

Transcriptome sequencing (TS): Dual DNA/RNA extraction is performed using the Qiagen Allprep DNA/RNA FFPE kit and quantitated using the Qubit 4.0 instrument. The Illumina TruSeq Stranded Total RNA Library Prep Gold kit is used to create whole transcriptome libraries. During library preparation, total RNA is depleted of ribosomal RNA (including mitochondrial ribosomal RNA), first and second strand cDNA is synthesized, A-tailed, adapters are ligated, and amplified using PCR. Library quality is validated using shallow MiSeq sequencing to assess mapping to ribosomal sequencing and transcriptomic regions. Transcriptome libraries are sequenced using 2x100 paired reads on an Illumina NovaSeq6000 system to a target depth of 100M PE reads (clusters) per sample.

Analysis: Analysis of WG and TS libraries proceeds through a validated pipeline after sequencing. FASTQ files called from the sequencing instrument are aligned using BWAmem for WG and STAR for TS. Quality control measures are performed on both the raw and aligned reads, which are evaluated by internal staff to ensure they meet minimum metrics. For WG, the aligned file is pre-processed, marking duplicates and normalizing the alignment, prior to variant calling. The processed BAM file is used to call somatic copy number variants, purity and ploidy, loss of heterozygosity (LOH), structural variants, somatic SNVs and indels, and tumour mutation burden. For WT, the BAM file is used to call fusion genes and evaluate percentile and gene expression outliers, as well as report immune infiltration. For RUO assays, the WG and WT results are then returned to the customer. In clinical assays, results from both WG and WT pipelines are passed to Djerba, which creates draft patient reports.

Clinical report: The clinical report details oncogenic copy number variants (CNVs), somatic single nucleotide variants (SNVs), insertion-deletions (indels), loss of heterozygosity (LOH), fusions, inferred cancer cell content (purity), and genome-wide ploidy with OncoKb annotation and evidence tiers for each variant class. Depending on customer needs, each report may also be issued with ‘research-use only’ results, which include immune inference analysis, microsatellite instability, or results from other tools with ongoing validation. Each component of the report is validated according to CAP, ACD and CLIA standards and new modules are adding according to stakeholder needs and community feedback. All clinical reports are reviewed by ACMG/CCMG board-certified geneticists before return to the customer.

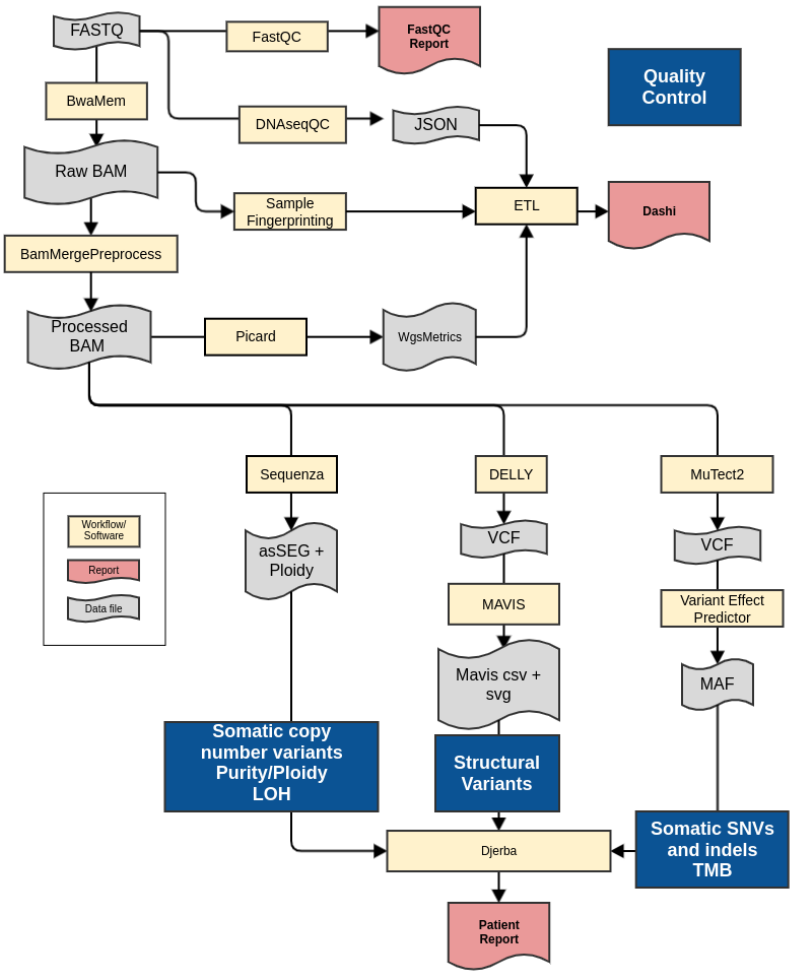

Figure 1: Validated whole genome (WG) pipeline. Starting from FASTQ reads, WG analysis proceeds through a number of analysis workflows and software. The dark blue boxes represent deliverables by the pipeline, which are used to ensure quality control and to create draft patient reports.

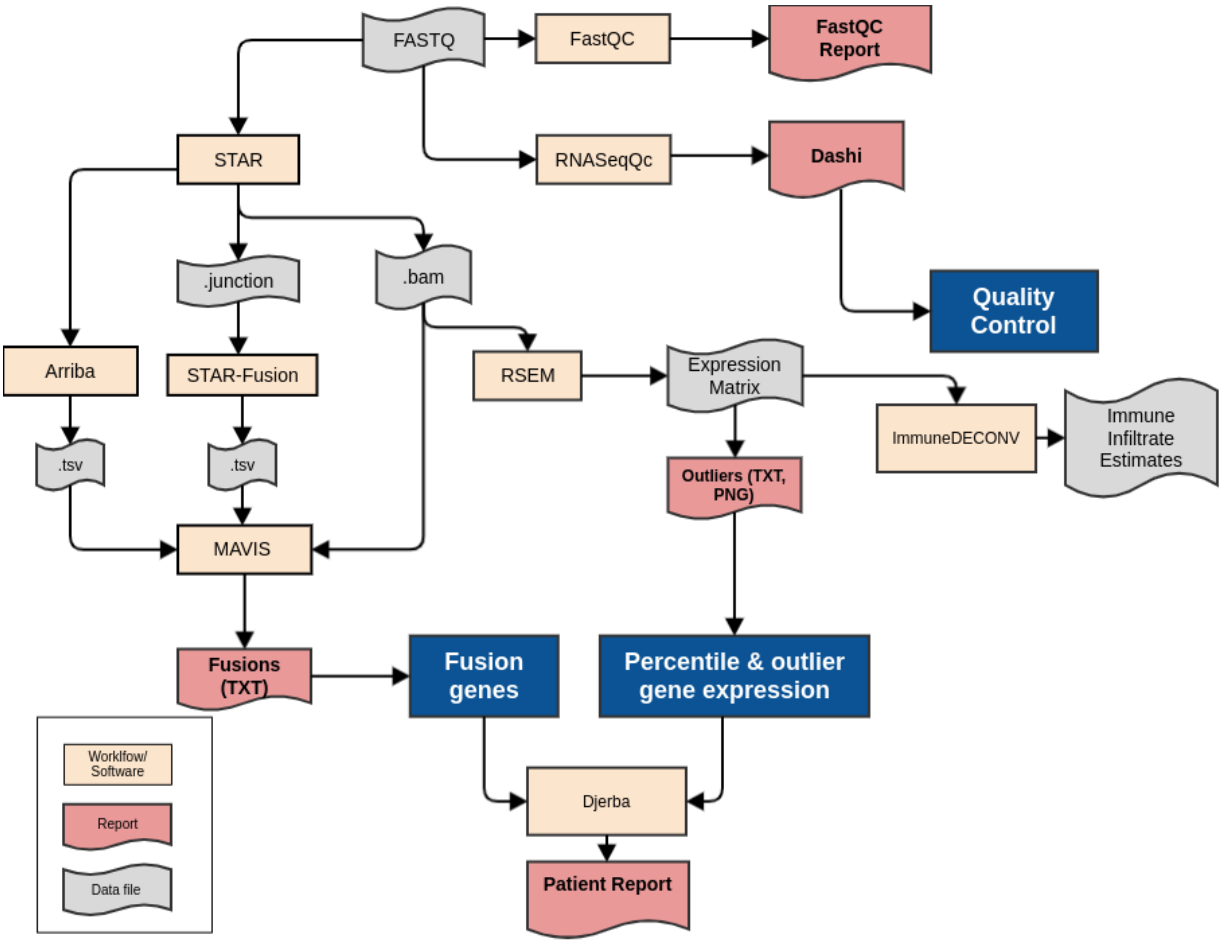

Figure 2: Validated whole transcriptome (TS) pipeline. As with the WG pipeline, TS begins with FASTQ reads that are analyzed by workflows and software to produce specific deliverables (dark blue boxes). The deliverables are used to ensure quality and create draft patient reports.