

**The Hospital for Sick Children  
Technology Assessment at SickKids (TASK)**

**APPENDIX**

**A MICROCOSTING AND COST-CONSEQUENCE ANALYSIS OF GENOMIC TESTING STRATEGIES (INCLUDING TRIOS) IN AUTISM SPECTRUM DISORDER: AN UPDATE**

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## **Appendix 1: Fact Sheet: Changes between the 2018 report update (2018-01) versus the 2016 report (2016-02.2)**

General for all platforms:

platforms in the 2018 report update: CMA, Illumina HiSeq<sup>®</sup> 2500, Illumina NextSeq<sup>®</sup> 550 and Illumina HiSeq X<sup>™</sup>; platforms in the 2016 report: CMA, Illumina HiSeq<sup>®</sup> 2500 and Illumina HiSeq X<sup>™</sup>:

- Overhead cost proportion: 22.3% vs. 23%
- No inflation adjustments were made for prices except labour from previous years for the 2018 report update; labour prices were adjusted by an annual 1.5% increase to 2017/2018. On the other hand, all prices were inflated for the 2016 report.

Exome and Genome analyses:

- Addition of filtration steps to analyze ~300-400 variants in order to flag variants of interest for both primary and secondary variants (impacts labour costing)

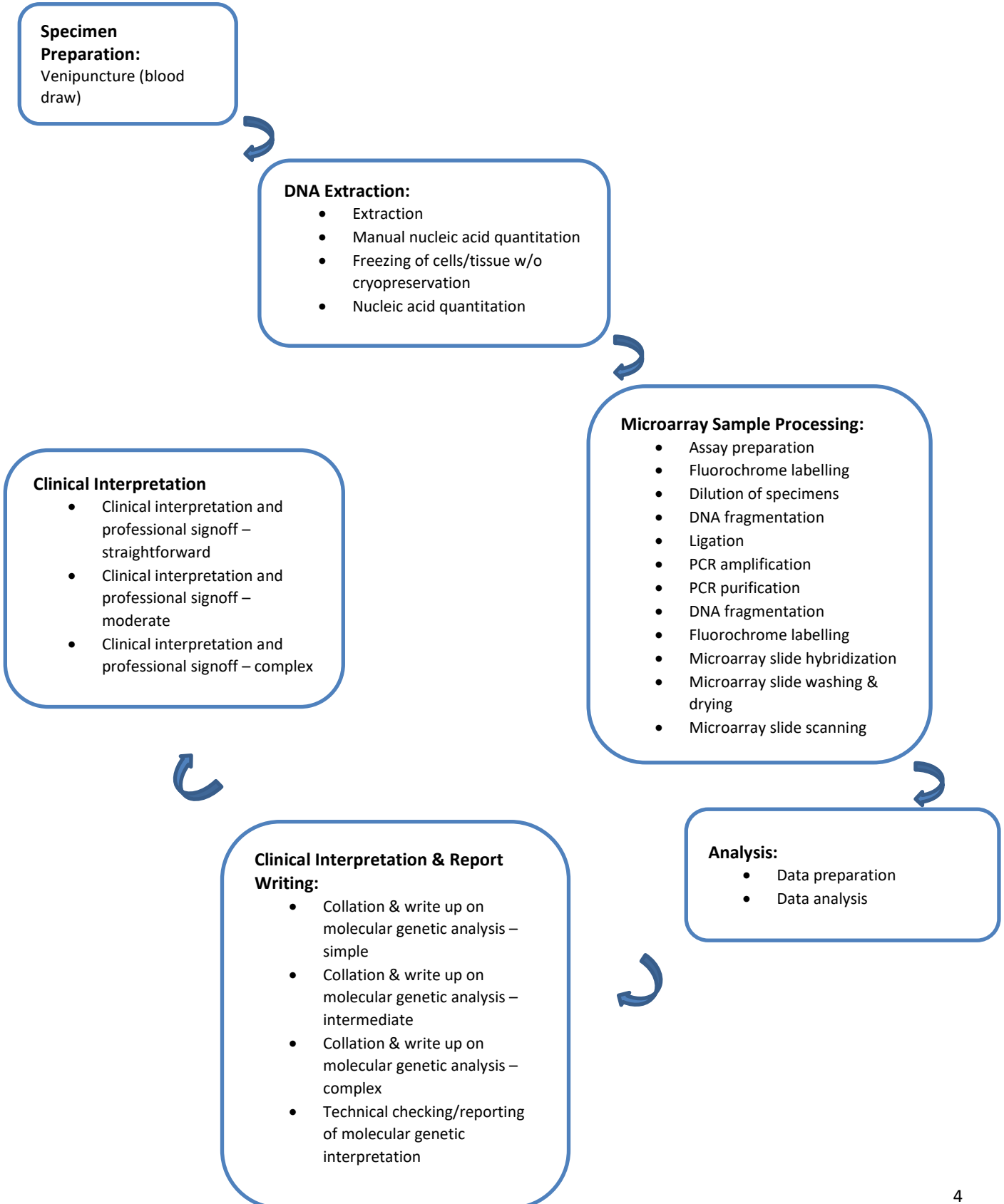
Whole Exome Sequencing:

- Addition of a new platform (NextSeq<sup>®</sup> 550)
- Exome output per month (NextSeq<sup>®</sup> 550): 80 (64-96)
- Exome output per month (HiSeq<sup>®</sup> 2500): 83 (70-95) vs. 87.5 (75-100)
- Bioinformatics
  - Pipeline: no change (Genome Analysis Toolkit (GATK))
  - Decrease of 1 step (temporary BAM files) for file storage
  - Additional 2 steps (extra step in recalibration; indel realignment) for computation use
  - Price per node: \$26,804 vs. \$9560

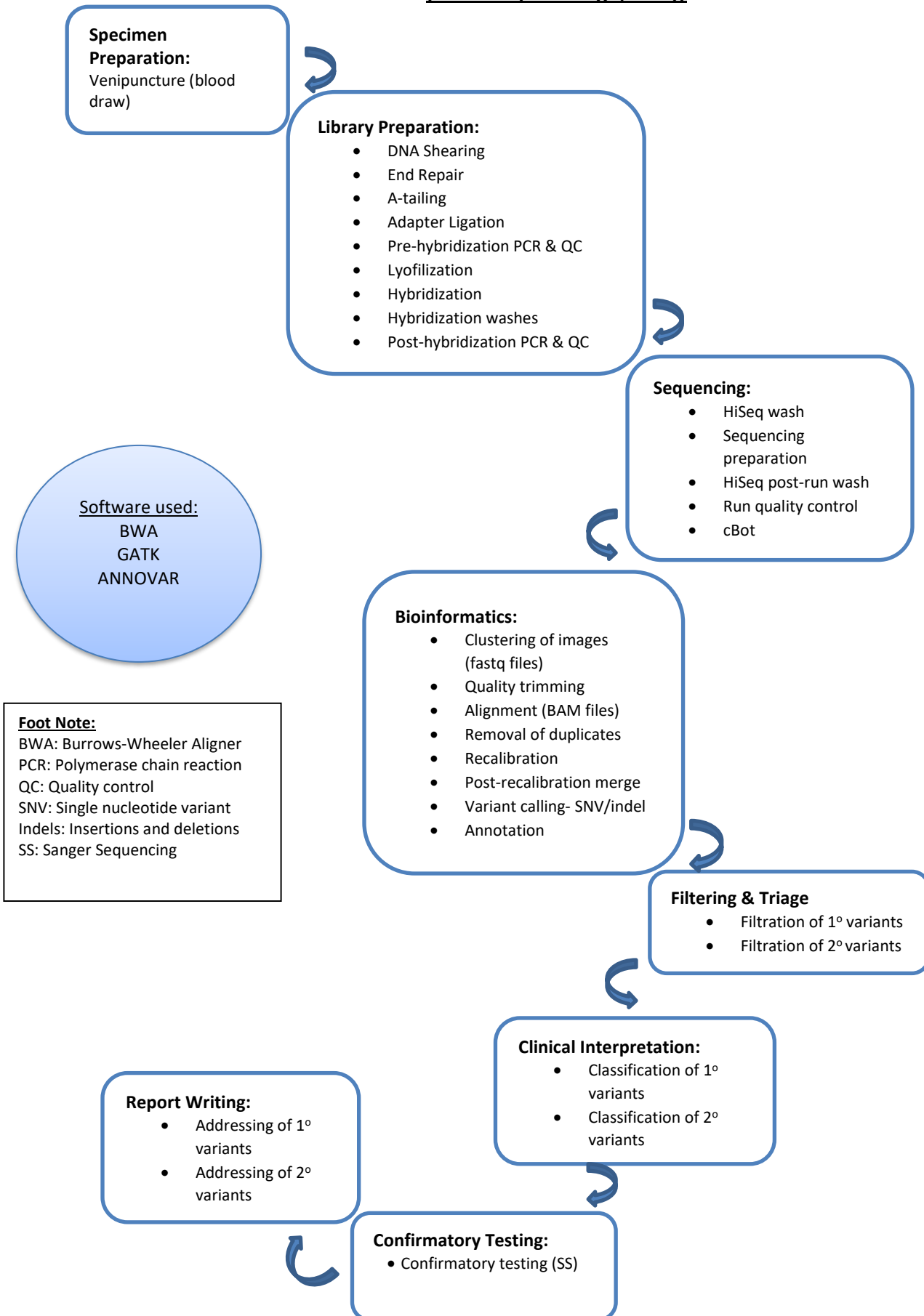
Whole Genome Sequencing:

- Elimination of platform HiSeq<sup>®</sup> 2500
- Inclusion of trio in addition to probands on the same platform (HiSeq X<sup>™</sup>)
  - Genome output per month 72 (64-96)
  - Proportion for follow-up testing (Sanger sequencing) is 10%
  - Small and large equipment volume: ½ of proband model
- Genome output per month 72 (64-96) vs. 150 (10% variability assigned)
- Bioinformatics
  - Pipeline: GATK vs. HAS
  - Additional 4 steps (recalibration; post-recalibration merge; CNV detection; SV detection) for computation use
  - Price per node: \$26,804 for all steps in 2018. In 2016, annotation was done on the old node (\$9560). Otherwise, same as 2018.

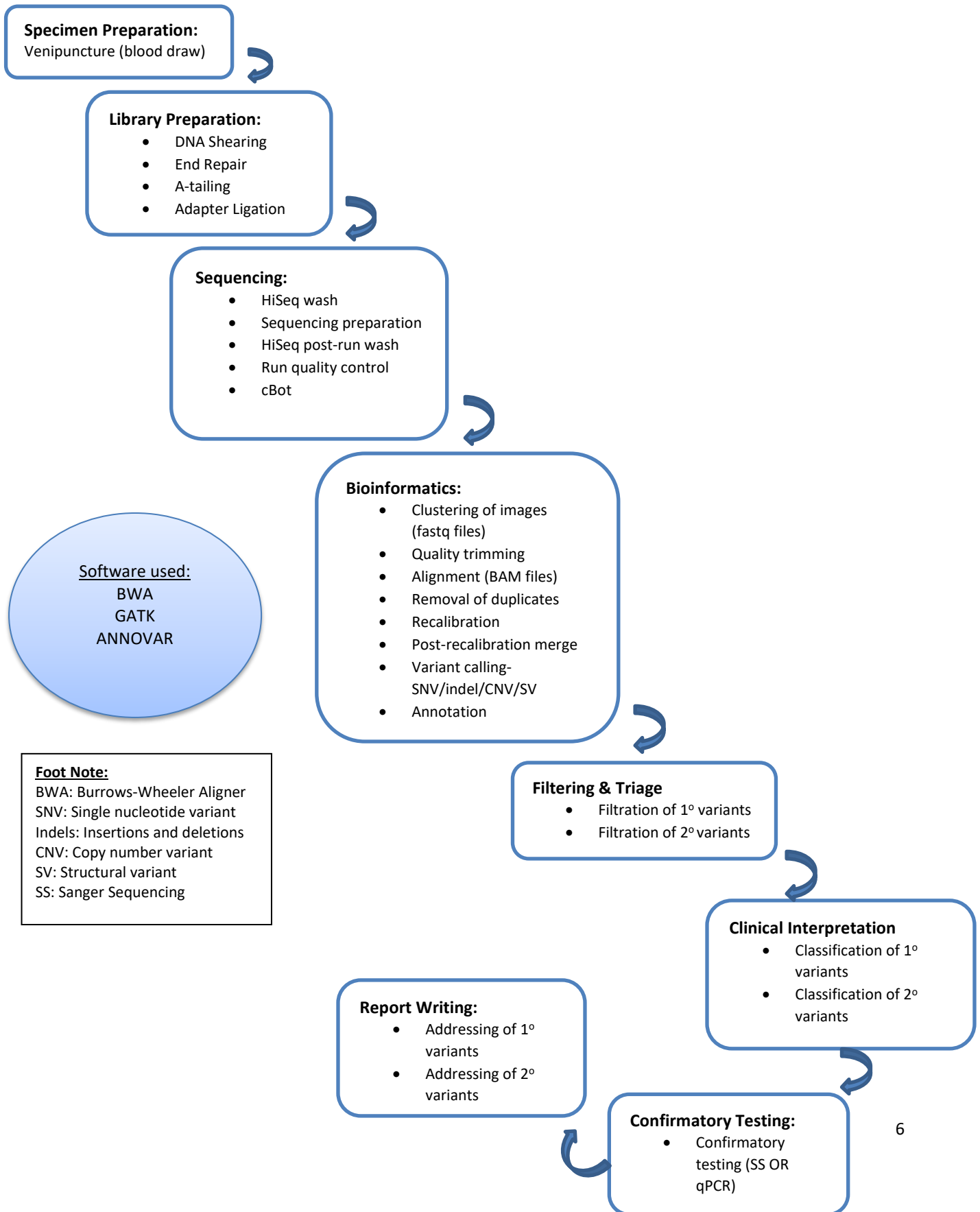
**Appendix 2: Chromosomal Microarray (CMA): Process Flow for Platform GeneChip® 3000Dx  
(Affymetrix (Santa Clara, USA))**



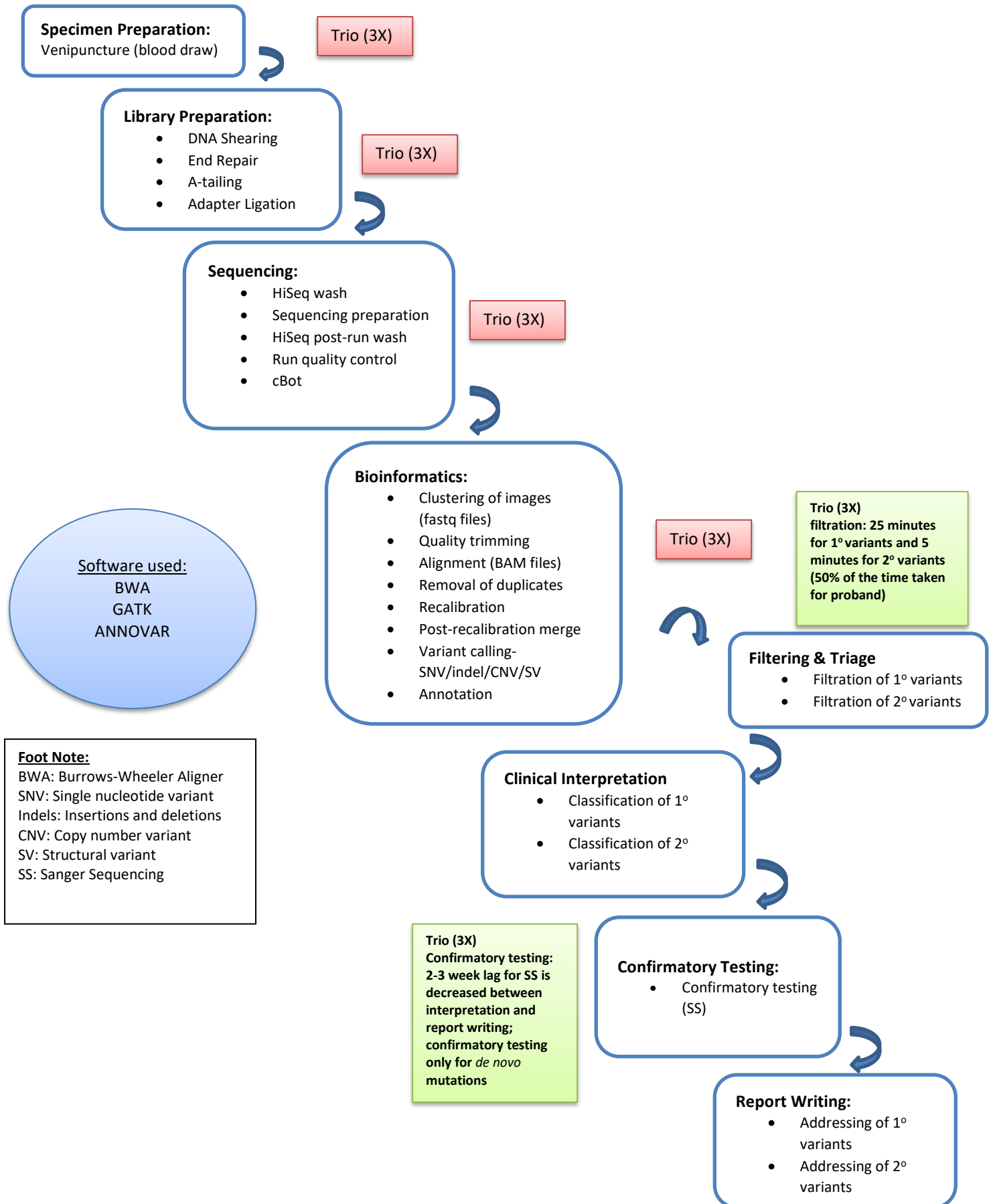
**Appendix 3: Whole Exome Sequencing (WES): Process Flow for Platform HiSeq® 2500  
(Illumina (San Diego, USA))**



**Appendix 4: Whole Genome Sequencing (WGS) (proband): Process Flow for Platform HiSeq X™ (Illumina (San Diego, USA))**



**Appendix 5: Whole Genome Sequencing (WGS) (trio): Process Flow for Platform HiSeq X™  
(Illumina (San Diego, USA))**



**Appendix 6. Resource use and unit prices for CMA inputs**

| Cost Items  | Quantity of Use per Sample |              |                    | Price (CAD/unit) |              |  |
|---|----------------------------|--------------|--------------------|------------------|--------------|--|
|   | Estimate                   | Range        | Source             | Estimate         | Range        | Source   |
| <b>Labour</b>   |                            |              |                    |                  |              |  |
| <b>Specimen Preparation (Units: minutes)</b>  |                            |              |                    |                  |              |  |
| 10120 Pediatric venipuncture  | 7.6                        | Not assigned | MIS Standards 2016 | Confidential     | -22% to +28% | SickKids wage information, 2014; 1.5% annual increase to 2017/18 |
| 11090 Packaging with testing documentation  | 1.0                        | Not assigned | MIS Standards 2016 | Confidential     | -22% to +28% | SickKids wage information, 2014; 1.5% annual increase to 2017/18 |
| 11500 Service recipient primary registration  | 1.8                        | Not assigned | MIS Standards 2016 | Confidential     | -20% to +20% | SickKids wage information, 2018                                  |
| 10360 Printing and sorting of specimen labels (per label)   | 0.4                        | Not assigned | MIS Standards 2016 | Confidential     | -20% to +20% | SickKids wage information, 2018                                  |
| 88550 Creation of family/service recipient folder (add sample to existing family folder, assign a new number and retrieve or make a new folder) (once per folder) | 5.0                        | Not assigned | MIS Standards 2016 | Confidential     | -20% to +20% | SickKids wage information, 2018                                  |
| 11090 Packaging with testing documentation  | 1.0                        | Not assigned | MIS Standards 2016 | Confidential     | -20% to +20% | SickKids wage information, 2018                                  |
| 11500 Service recipient limited registration  | 1.8                        | Not assigned | MIS Standards 2016 | Confidential     | -20% to +20% | SickKids wage information, 2018                                  |
| <b>DNA Extraction (Units: minutes)</b>  |                            |              |                    |                  |              |  |
| 85340 Extraction using an automated kit (once per specimen)   | 2.0                        | Not assigned | MIS Standards 2016 | Confidential     | -20% to +20% | SickKids wage information, 2018                                  |
| 85360 Manual nucleic acid quantitation (once per specimen)  | 5.0                        | Not assigned | MIS Standards 2016 | Confidential     | -20% to +20% | SickKids wage information, 2018                                  |
| 84510 Freezing of cells/tissue without cryopreservation   | 9.0                        | Not assigned | MIS Standards 2016 | Confidential     | -20% to +20% | SickKids wage information, 2018                                  |



| Cost Items   | Quantity of Use per Sample |              |                             | Price (CAD/unit) |              |                                 |
|--|----------------------------|--------------|-----------------------------|------------------|--------------|---------------------------------|
|  | Estimate                   | Range        | Source                      | Estimate         | Range        | Source                          |
| 85370 Nucleic acid quantitation using spectrophotometer with sample retention tech (once per specimen) | 1.0                        | Not assigned | MIS Standards 2016          | Confidential     | -20% to +20% | SickKids wage information, 2018 |
| <b>Microarray sample processing (Units: minutes)</b>   |                            |              |                             |                  |              |                                 |
| 85500 Assay preparation - manual worksheet prep (3 units for each assay and 1 per specimen)            | 2.0                        | Not assigned | MIS Standards 2016          | Confidential     | -20% to +20% | SickKids wage information, 2018 |
| 83440 Fluorochrome labelling without dye swap  | 4.0                        | Not assigned | MIS Standards 2016          | Confidential     | -20% to +20% | SickKids wage information, 2018 |
| 85560 Dilution of specimens (at the assay stage only, once per specimen)                               | 2.0                        | Not assigned | MIS Standards 2016          | Confidential     | -20% to +20% | SickKids wage information, 2018 |
| DNA Fragmentation, by Restriction Enzyme Digestion   | 2.3                        | Not assigned | Cytogenetics lab specialist | Confidential     | -20% to +20% | SickKids wage information, 2018 |
| Ligation   | 1.5                        | Not assigned | Cytogenetics lab specialist | Confidential     | -20% to +20% | SickKids wage information, 2018 |
| PCR amplification  | 2.3                        | Not assigned | Cytogenetics lab specialist | Confidential     | -20% to +20% | SickKids wage information, 2018 |
| PCR purification by magnetic beads (once per sample)   | 12.4                       | Not assigned | Cytogenetics lab specialist | Confidential     | -20% to +20% | SickKids wage information, 2018 |
| DNA Fragmentation-by Restriction Enzyme Digestion  | 2.3                        | Not assigned | Cytogenetics lab specialist | Confidential     | -20% to +20% | SickKids wage information, 2018 |
| Fluorochrome labelling without dye swap.   | 1.1                        | Not assigned | Cytogenetics lab specialist | Confidential     | -20% to +20% | SickKids wage information, 2018 |
| Microarray Slide Hybridization   | 4.1                        | Not assigned | Cytogenetics lab specialist | Confidential     | -20% to +20% | SickKids wage information, 2018 |
| Microarray Slide Washing and Drying - Automated  | 8.0                        | Not assigned | Cytogenetics lab specialist | Confidential     | -20% to +20% | SickKids wage information, 2018 |
| Microarray slide scanning - Affymetrix scanner (once per slide)  | 10.0                       | Not assigned | Cytogenetics lab specialist | Confidential     | -20% to +20% | SickKids wage information, 2018 |
| <b>Analysis (Units: minutes)</b>   |                            |              |                             |                  |              |                                 |
| Data preparation (includes transfer of data images)  | 8.0                        | Not assigned | Cytogenetics lab specialist | Confidential     | -20% to +20% | SickKids wage information, 2018 |

| Cost Items  | Quantity of Use per Sample |                |                           | Price (CAD/unit) |                       |   |
|---|----------------------------|----------------|---------------------------|------------------|-----------------------|---|
|   | Estimate                   | Range          | Source                    | Estimate         | Range                 | Source  |
| 83550 Data analysis (includes review and analysis of each chromosome, prep prelim report, once per service recipient) | 12.0                       | Not assigned   | Lab technician            | Confidential     | -20% to +20%          | SickKids wage information, 2018                                     |
| <b>Clinical interpretation and report writing (Units: minutes)</b>  |                            |                |                           |                  |                       |   |
| 87760 Collation and write up on molecular genetic analysis - simple (60% of cases)                                    | 2.0                        | Not assigned   | MIS Standards 2016        | Confidential     | -20% to +20%          | SickKids wage information, 2018                                     |
| 87760 Collation and write up on molecular genetic analysis - intermediate (20% of cases)                              | 10.0                       | Not assigned   | MIS Standards 2016        | Confidential     | -23% to +28%          | Lab director, 2014; 1.5% annual increase to 2017/18                 |
| 87760 Collation and write up on molecular genetic analysis - complex (20% of cases)                                   | 50.0                       | Not assigned   | MIS Standards 2016        | Confidential     | -23% to +28%          | Lab director, 2014; 1.5% annual increase to 2017/18                 |
| 87770 Technical checking/reporting of molecular genetic interpretation (30% of cases)                                 | 5.0                        | Not assigned   | Cytogenetics lab director | Confidential     | -31% to +15%          | Informatics facility manager, 2014; 1.5% annual increase to 2017/18 |
| <b>Clinical interpretation (Units: minutes)</b>   |                            |                |                           |                  |                       |   |
| 87780 Clinical interpretation and professional signoff (80% of cases, 10 minutes per case)                            | 8.0                        | Not assigned   | Cytogenetics lab director | Confidential     | -20% to +20%          | Public sector disclosure 2012; 1.5% annual increase to 2017/18      |
| 87780 Clinical interpretation and professional signoff (15% of cases, 20 minutes per case)                            | 3.0                        | Not assigned   | Cytogenetics lab director | Confidential     | -20% to +20%          | Public sector disclosure 2012; 1.5% annual increase to 2017/18      |
| 87780 Clinical interpretation and professional signoff (5% of cases, 60 minutes per case)                             | 3.0                        | Not assigned   | Cytogenetics lab director | Confidential     | -20% to +20%          | Public sector disclosure 2012; 1.5% annual increase to 2017/18      |
| <b>Large Equipment</b>  |                            |                |                           |                  |                       |   |
| Affymetrix 1 GeneChip 3000Dx, 2 Fluidics stations, 1 hybridization oven   | 2/all tests                | Not applicable | Lab manager               | 398,152          | 358,336.80-437,967.20 | Affymetrix sales representative, 2018                               |
| 1-year service contract   | 1/all tests                | Not applicable | Affymetrix representative | 89,277.20        |                       | Affymetrix sales representative, 2018                               |

| Cost Items  | Quantity of Use per Sample |              |                | Price (CAD/unit) |                       |                                       |
|---|----------------------------|--------------|----------------|------------------|-----------------------|---------------------------------------|
|   | Estimate                   | Range        | Source         | Estimate         | Range                 | Source                                |
|   |                            |              |                |                  | 80,349.48 – 98,204.92 |                                       |
| <b>Supplies</b>   |                            |              |                |                  |                       |                                       |
| Shipping & handling   | 1                          | Not assigned | FedEx          | 37.61            | 33.80 – 41.40         | FedEx, 2018                           |
| Microarray slide & reagents per patient   | 1                          | Not assigned | Cytogeneticist | Confidential     | -10% to +10%          | Affymetrix sales representative, 2018 |
| <b>Follow-up (proportion of cases)</b>  |                            |              |                |                  |                       |                                       |
| Note: Price for FISH and qPCR tests includes three tests, proband and two parents |                            |              |                |                  |                       |                                       |
| FISH follow-up (proband and two parents)  | 0.1                        | Not assigned | Cytogeneticist | 680.00           | 578.04 – 720.50       | Cytogenetics lab director, 2018       |
| qPCR follow-up (proband and two parents)  | 0.05                       | Not assigned | Cytogeneticist | 223.90           | 201.51 – 246.29       | Expert opinion, 2018                  |

Abbreviations: CMA, Chromosomal Microarray Analysis; PCR, Polymerase chain reaction; MIS, Management Information Systems; qPCR, Real-time polymerase chain reaction; FISH, Fluorescence in situ hybridization.

**Appendix 7. Resource use and unit prices for WES inputs, Illumina HiSeq® 2500**

| Cost Items  | Quantity of Use per Sample |              |                    | Price (CAD \$/unit) |              |  |
|---|----------------------------|--------------|--------------------|---------------------|--------------|--|
|   | Estimate                   | Range        | Source             | Estimate            | Range        | Source   |
| <b>Labour</b>   |                            |              |                    |                     |              |  |
| <b>Specimen Preparation (Units: minutes)</b>  |                            |              |                    |                     |              |  |
| 10120 Pediatric venipuncture  | 7.6                        | Not assigned | MIS Standards 2016 | Confidential        | -22% to +28% | SickKids wage information, 2014; 1.5% annual increase to 2017/18 |
| 11090 Packaging with testing documentation  | 1.0                        | Not assigned | MIS Standards 2016 | Confidential        | -22% to +28% | SickKids wage information, 2014; 1.5% annual increase to 2017/18 |
| 11500 Service recipient primary registration  | 1.8                        | Not assigned | MIS Standards 2016 | Confidential        | -20% to +20% | SickKids wage information, 2018                                  |
| 10360 Printing and sorting of specimen labels (per label)   | 0.4                        | Not assigned | MIS Standards 2016 | Confidential        | -20% to +20% | SickKids wage information, 2018                                  |
| 88550 Creation of family/service recipient folder (add sample to existing family folder, assign a new number and retrieve or make a new folder) (once per folder) | 5.0                        | Not assigned | MIS Standards 2016 | Confidential        | -20% to +20% | SickKids wage information, 2018                                  |
| 11090 Packaging with testing documentation  | 1.0                        | Not assigned | MIS Standards 2016 | Confidential        | -20% to +20% | SickKids wage information, 2018                                  |
| 11500 Service recipient limited registration  | 1.8                        | Not assigned | MIS Standards 2016 | Confidential        | -20% to +20% | SickKids wage information, 2018                                  |
| <b>Library preparation (Units: minutes)</b>   |                            |              |                    |                     |              |  |
| Note: Total time per task was divided by the number of samples per batch. Resource use estimate was based on 8 samples per batch.                                 |                            |              |                    |                     |              |  |
| DNA quantification (20 min)   | 2.5                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| Pre-prep reagents (20 min)  | 2.5                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| Shearing (20 min)   | 2.5                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| Purification (40 min)   | 5.0                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| End repair (40 min)   | 5.0                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| A-tailing (40 min)  | 5.0                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| Adapter ligation (45 min)   | 5.6                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| Pre-hybridization PCR (45 min)  | 5.6                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |

| Cost Items   | Quantity of Use per Sample |                 |                                   | Price (CAD \$/unit) |              |                                   |
|--|----------------------------|-----------------|-----------------------------------|---------------------|--------------|-----------------------------------|
|  | Estimate                   | Range           | Source                            | Estimate            | Range        | Source                            |
| Pre-hybridization QC (60 min)  | 7.5                        | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| Lyophilization (20 min)  | 2.5                        | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| Hybridization (30 min)   | 3.8                        | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| Hybridization washes (150 min)   | 18.8                       | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| Post-Hybridization PCR (40 min)  | 5.0                        | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| Post Hybridization QC (120 min)  | 15.0                       | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| <b>Sequencing (Units: minutes)</b>   |                            |                 |                                   |                     |              |                                   |
| Note: Total time per task was divided by the number of samples per lane. Resource use estimate was based on 8 samples per lane.  |                            |                 |                                   |                     |              |                                   |
| HiSeq wash (30 min)  | 3.8                        | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| Sequencing prep (30 min)   | 3.8                        | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| HiSeq post-run wash (45 min)   | 5.6                        | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| Run quality control (15 min)   | 1.9                        | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| cBot (30 min)  | 3.8                        | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| <b>Bioinformatics (Units: minutes)</b>   |                            |                 |                                   |                     |              |                                   |
| Note: Resource use estimate was based on average output, 83 exomes per month. The lower bound was based on 95 exomes and the upper bound was based on to 70 exomes.  |                            |                 |                                   |                     |              |                                   |
| Variant calling (1 FTE hour for 70-95 exomes a month)  | 101.2                      | 88.4-120.0      | TCAG Bioinformatics manager       | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |
| Annotation (0.25 FTE hour for 70-95 exomes a month)  | 25.3                       | 22.1-30.0       | TCAG Bioinformatics manager       | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |
| <b>Bioinformatics Maintenance (Units: minutes)</b>   |                            |                 |                                   |                     |              |                                   |
| Note: One full time equivalent (FTE) hour to support one node per year was assumed. Resource use in minutes was calculated by multiplying the time to complete a job by the time needed to support the required number of nodes per job. For example, if a job required 0.5 hours and 3 nodes, then the resource use for the job is calculated as follows: 60 FTE minutes in an hour per year/(365*24 hours in a year)*3 hours to support 3 nodes*0.5 hours (i.e. number of minutes per year to support a 30 minute job on 3 nodes). The lower bound of resource use was based on 75% of the computation time and the upper bound was based on 125% of computation time. |                            |                 |                                   |                     |              |                                   |
| Alignment<br>0.5 hour (range: 0.4-0.6) 1 node  | 0.00342                    | 0.00274-0.00411 | TCAG Bioinformatics manager, 2018 | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |
| Mark Duplicates - PICARD<br>1 hour (range: 0.75-1.25) 1/20 nodes   | 0.00034                    | 0.00026-0.00043 | TCAG Bioinformatics manager, 2018 | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |
| Recalibration- GATK (step 1)<br>1.5 hours (range: 4-6) 1/20 nodes  | 0.00051                    | 0.00043-0.00060 | TCAG Bioinformatics manager, 2018 | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |

| Cost Items  | Quantity of Use per Sample |                    |   | Price (CAD \$/unit) |              |                                       |
|---|----------------------------|--------------------|---|---------------------|--------------|---------------------------------------|
|   | Estimate                   | Range              | Source                                    | Estimate            | Range        | Source                                |
| Recalibration – GATK (step 2)<br>0.5 hour (range: 0.25-0.75) 2 nodes  | 0.00685                    | 0.00342-0.0103     | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| Post-recalibration merge – GATK<br>0.5 hour (range: 0.25-0.75) 1/20 nodes   | 0.000171                   | 0.0000856-0.000257 | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| Indel Realignment (GATK)<br>0.5 hour (range: 0.25-0.75) 2 nodes   | 0.01                       | 0.00342-0.0103     | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| SNV/indel variant calling - (GATK haplotype caller)<br>0.33 hour (range: 0.3-0.37) 2 nodes  | 0.00456                    | 0.00411-0.00507    | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| Annotation (ANNOVAR)<br>0.5 hour (range: 0.25-0.75) 0.2 nodes   | 0.00069                    | 0.000342-0.00103   | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| <b>Filtering &amp; Triage (Units: minutes)</b>  |                            |                    |   |                     |              |                                       |
| Filtration of primary variants  | 55                         | Not assigned       | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20% | Public Sector Salary Disclosure, 2017 |
| Filtration of secondary variants  | 5                          | Not assigned       | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20% | Public Sector Salary Disclosure, 2017 |
| <b>Clinical Interpretation (Units: minutes)</b>   |                            |                    |   |                     |              |                                       |
| Clinical classification of primary variants<br>15 minutes if no variants are found, otherwise 30 minutes per variant. On average, 2 variants are found (range: 0-4) | 60                         | 15-120             | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20% | Public Sector Salary Disclosure, 2017 |
| Clinical classification of secondary variants<br>30 minutes per case (range: 20-40), found in 4% of cases (range: 3-5%)   | 1.2                        | 0.6-2.0            | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20% | Public Sector Salary Disclosure, 2017 |
| <b>Report Writing (Units: minutes)</b>  |                            |                    |   |                     |              |                                       |

| Cost Items  | Quantity of Use per Sample |                |   | Price (CAD \$/unit) |               |   |
|---|----------------------------|----------------|---|---------------------|---------------|---|
|   | Estimate                   | Range          | Source                                    | Estimate            | Range         | Source  |
| Addressing primary variants<br>15 minutes base plus 15 minutes per variant. On average, 2 variants are found (range: 0-4) | 45                         | 15-75          | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20%  | Public Sector Salary Disclosure, 2017         |
| Addressing secondary variants<br>30 minutes per case (range: 20-40), found in 4% of cases (range: 3-5%)                   | 1.2                        | 0.6-2.0        | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20%  | Public Sector Salary Disclosure, 2017         |
| <b>Large Equipment</b>  |                            |                |   |                     |               |   |
| Illumina HiSeq® 2500  | 1/all tests                | Not applicable | TCAG lab manager                          | 750000              | 700000-800000 | TCAG lab manager, 2015                        |
| 1-year service contract   | 1/all tests                | Not applicable | TCAG lab manager                          | 75000               | 50000-82500   | TCAG lab manager, 2015                        |
| Agilent BioAnalyzer/Tape station  | 1/all tests                | Not applicable | Agilent representative                    | 38500               | 34000-43000   | Agilent representative/TCAG lab manager, 2015 |
| <b>Small Equipment</b>  |                            |                |   |                     |               |   |
| Tube microcentrifuge  | 1/all tests                | Not applicable | TCAG lab manager                          | 2250                | 2000-2500     | TCAG lab manager, 2014                        |
| Plate microcentrifuge   | 1/all tests                | Not applicable | TCAG lab manager                          | 5000                | 4500-5500     | TCAG lab manager, 2014                        |
| Thermomixer   | 1/all tests                | Not applicable | TCAG lab manager                          | 5000                | 4500-5500     | TCAG lab manager, 2014                        |
| Vortex  | 1/all tests                | Not applicable | TCAG lab manager                          | 450                 | 400-500       | TCAG lab manager, 2014                        |
| Pipette sets  | 2/all tests                | Not applicable | TCAG lab manager                          | 1600                | 1400-2000     | TCAG lab manager, 2014                        |
| Magnet particle concentrator for tubes  | 1/all tests                | Not applicable | TCAG lab manager                          | 700                 | 630-770       | TCAG lab manager, 2014                        |
| Thermocyclers   | 2/all tests                | Not applicable | TCAG lab manager                          | 3000                | 2700-3300     | TCAG lab manager, 2014                        |
| <b>Supplies</b>   |                            |                |   |                     |               |   |
| Shipping & handling   | 1                          | Not assigned   | TCAG lab Manager                          | 37.61               | 33.80-41.40   | FedEx, 2018                                   |
| SureSelect Baits (96 reactions)   | 1                          | Not assigned   | TCAG lab manager                          | 195                 | 175.50-214.50 | TCAG lab manager, 2018                        |
| SureSelect Library prep (96 reactions)  | 1                          | Not assigned   | TCAG lab manager                          | 22.50               | 20.30-24.80   | TCAG lab manager, 2018                        |
| Other library prep consumables  | 1                          | Not assigned   | TCAG lab manager                          | 70.0                | 63.00-77.00   | TCAG lab manager, 2018                        |
| Sequencing reagents (8 samples per lane)  | 1                          | Not assigned   | TCAG lab manager                          | 337.50              | 303.80-371.30 | TCAG lab manager 2018                         |
| <b>Follow-up (proportion of cases)</b>  |                            |                |   |                     |               |   |

| Cost Items   | Quantity of Use per Sample |              |   | Price (CAD \$/unit) |             |   |
|--|----------------------------|--------------|---|---------------------|-------------|---|
|  | Estimate                   | Range        | Source                                    | Estimate            | Range       | Source  |
| Sanger sequencing (two tests per person, proband and two parents)  | 0.5                        | Not assigned | DPLM Associate director, Genomic Analysis | 53.33               | 48.00-58.70 | DPLM Associate director, Genomic Analysis, 2018 |
| <b>Bioinformatics</b>  |                            |              |   |                     |             |   |
| <b>Bioinformatics File Storage (Units: GB per year)</b>  |                            |              |   |                     |             |   |
| Notes: 1. Resource use volume was calculated by multiplying the file size by the storage time (in years).<br>2. Price was given in dollars (\$) per GB per year.   |                            |              |   |                     |             |   |
| trimmed fastq<br>file size: 9 GB; storage time: 9 months (range: 6-12)   | 6.75                       | 4.5-9.0      | TCAG Bioinformatics manager               | 0.4                 | 0.36-0.44   | TCAG Bioinformatics manager, 2014               |
| final rem-dup, recalibrated, locally re-aligned BAM file<br>file size: 9 GB; storage time: 9 months (range: 6-12)  | 4.5                        | 3.0-6.0      | TCAG Bioinformatics manager               | 0.4                 | 0.36-0.44   | TCAG Bioinformatics manager, 2014               |
| <b>Bioinformatics Computation Use (Units: CPU time per hour)</b>   |                            |              |   |                     |             |   |
| Notes: 1. Resource use volume was calculated by multiplying the number of jobs by the number of cores per job and time (hours) required to complete a job. 25% (range 0 to 50%) was added to account for additional processing time to avoid operating at full capacity.<br>2. Scalar Decisions, Inc. quote of \$26,804 per node (20 cores) or \$1340.20 per core (including warranty) was used. A straight line depreciation method over 5 years was applied and the price per CPU per year was calculated to be \$5360.80. This price was converted to price per CPU per hour by dividing \$5360.80 by (365*24). |                            |              |   |                     |             |   |
| Alignment<br>1 job, 54 GB RAM per job, 19 core per job, 0.5 hour to complete, >>1 node required  | 11.875                     | 9.5-14.25    | TCAG Bioinformatics manager               | 0.612               | 0.551-0.673 | Scalar quote, 2016                              |
| Mark Duplicates – PICARD<br>1 job, 5 GB RAM per job, 2 cores per job, 1 hour to complete, >> 1/20 node required  | 2.5                        | 2-3          | TCAG Bioinformatics manager               | 0.612               | 0.551-0.673 | Scalar quote, 2016                              |
| Recalibration- GATK (step 1)<br>1 job, 30 GB RAM per job, 1 core per job, 1.5 hours to complete, >>1/20 node required  | 1.875                      | 1.5-2.25     | TCAG Bioinformatics manager               | 0.612               | 0.551-0.673 | Scalar quote, 2016                              |



| Cost Items   | Quantity of Use per Sample |            |                             | Price (CAD \$/unit) |             |                    |
|--|----------------------------|------------|-----------------------------|---------------------|-------------|--------------------|
|  | Estimate                   | Range      | Source                      | Estimate            | Range       | Source             |
| Recalibration – GATK (step 2)<br>23 jobs, 30 GB RAM per job, 1 core per job, 0.5 hour to complete, >>2 nodes required                      | 14.375                     | 11.5-17.25 | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | Scalar quote, 2016 |
| Post-recalibration merge - (GATK)<br>1 job, 10 GB RAM per job, 2 core per job, 0.5 hour to complete, >>1/20 node required                  | 1.25                       | 1-1.5      | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | Scalar quote, 2016 |
| Indel Realignment (GATK)<br>23 jobs, 10 GB RAM per job, 2 core per job, 0.5 hour to complete, >>2 nodes required                           | 28.75                      | 23-34.5    | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | Scalar quote, 2016 |
| SNV/indel variant calling - GATK haplotype caller<br>23 jobs, 55 GB RAM per job, 1 core per job, 0.33 hour to complete, >>2 nodes required | 9.58                       | 7.67-11.5  | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | Scalar quote, 2016 |
| Annotation (ANNOVAR)<br>1 job, 32 GB RAM per job, 8 cores per job, 0.5 hour to complete, >>1/5 node required                               | 5                          | 4-6        | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | Scalar quote, 2016 |

Abbreviations: WES, Whole Exome Sequencing; TCAG, The Centre for Applied Genomics; DPLM, The Department of Paediatric Laboratory Medicine; FTE, Full-time equivalent; MIS, Management Information Systems; GATK, Genome Analysis Toolkit; SNV, Single nucleotide variant.

**Appendix 8. Resource use and unit prices for WES inputs, Illumina NextSeq® 550**

| Cost Items  | Quantity of Use per Sample |              |                    | Price (CAD \$/unit) |              |  |
|---|----------------------------|--------------|--------------------|---------------------|--------------|--|
|   | Estimate                   | Range        | Source             | Estimate            | Range        | Source   |
| <b>Labour</b>   |                            |              |                    |                     |              |  |
| <b>Specimen Preparation (Units: minutes)</b>  |                            |              |                    |                     |              |  |
| 10120 Pediatric venipuncture  | 7.6                        | Not assigned | MIS Standards 2016 | Confidential        | -22% to +28% | SickKids wage information, 2014; 1.5% annual increase to 2017/18 |
| 11090 Packaging with testing documentation  | 1.0                        | Not assigned | MIS Standards 2016 | Confidential        | -22% to +28% | SickKids wage information, 2014; 1.5% annual increase to 2017/18 |
| 11500 Service recipient primary registration  | 1.8                        | Not assigned | MIS Standards 2016 | Confidential        | -20% to +20% | SickKids wage information, 2018                                  |
| 10360 Printing and sorting of specimen labels (per label)   | 0.4                        | Not assigned | MIS Standards 2016 | Confidential        | -20% to +20% | SickKids wage information, 2018                                  |
| 88550 Creation of family/service recipient folder (add sample to existing family folder, assign a new number and retrieve or make a new folder) (once per folder) | 5.0                        | Not assigned | MIS Standards 2016 | Confidential        | -20% to +20% | SickKids wage information, 2018                                  |
| 11090 Packaging with testing documentation  | 1.0                        | Not assigned | MIS Standards 2016 | Confidential        | -20% to +20% | SickKids wage information, 2018                                  |
| 11500 Service recipient limited registration  | 1.8                        | Not assigned | MIS Standards 2016 | Confidential        | -20% to +20% | SickKids wage information, 2018                                  |
| <b>Library preparation (Units: minutes)</b>   |                            |              |                    |                     |              |  |
| Note: Total time per task was divided by the number of samples per batch. Resource use estimate was based on 8 samples per batch.                                 |                            |              |                    |                     |              |  |
| DNA quantification (20 min)   | 2.5                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| Pre-prep reagents (20 min)  | 2.5                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| Shearing (20 min)   | 2.5                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| Purification (40 min)   | 5.0                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| End repair (40 min)   | 5.0                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| A-tailing (40 min)  | 5.0                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| Adapter ligation (45 min)   | 5.6                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| Pre-hybridization PCR (45 min)  | 5.6                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |

| Cost Items   | Quantity of Use per Sample |                 |                                   | Price (CAD \$/unit) |              |                                   |
|--|----------------------------|-----------------|-----------------------------------|---------------------|--------------|-----------------------------------|
|  | Estimate                   | Range           | Source                            | Estimate            | Range        | Source                            |
| Pre-hybridization QC (60 min)  | 7.5                        | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| Lyophilization (20 min)  | 2.5                        | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| Hybridization (30 min)   | 3.8                        | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| Hybridization washes (150 min)   | 18.8                       | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| Post-Hybridization PCR (40 min)  | 5.0                        | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| Post Hybridization QC (120 min)  | 15.0                       | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| <b>Sequencing (Units: minutes)</b>   |                            |                 |                                   |                     |              |                                   |
| Note: Total time per task was divided by the number of samples per lane. Resource use estimate was based on 8 samples per lane.  |                            |                 |                                   |                     |              |                                   |
| Sequencing prep (30 min)   | 3.8                        | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| Run quality control (15 min)   | 1.9                        | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| <b>Bioinformatics (Units: minutes)</b>   |                            |                 |                                   |                     |              |                                   |
| Note: Resource use estimate was based on the average output, 80 exomes per month. The lower bound was based on 96 exomes and the upper bound was based on 64 exomes.   |                            |                 |                                   |                     |              |                                   |
| Variant calling (1 FTE hour for 64-96 exomes a month)  | 105                        | 87.5-131.25     | TCAG Bioinformatics manager       | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |
| Annotation (0.25 FTE hour for 64-96 exomes a month)  | 26.25                      | 21.875-32.813   | TCAG Bioinformatics manager       | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |
| <b>Bioinformatics Maintenance (Units: minutes)</b>   |                            |                 |                                   |                     |              |                                   |
| Note: One full time equivalent (FTE) hour to support one node per year was assumed. Resource use in minutes was calculated by multiplying the time to complete a job by the time needed to support the required number of nodes per job. For example, if a job required 0.5 hours and 3 nodes, then the resource use for the job is calculated as follows: 60 FTE minutes in an hour per year/ (365*24 hours in a year) *3 hours to support 3 nodes*0.5 hours (i.e. number of minutes per year to support a 30 minute job on 3 nodes). The lower bound of resource use was based on 75% of the computation time and the upper bound was based on 125% of computation time. |                            |                 |                                   |                     |              |                                   |
| Alignment<br>0.5 hour (range: 0.4-0.6) 1 node  | 0.00342                    | 0.00274-0.00411 | TCAG Bioinformatics manager, 2018 | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |
| Mark Duplicates - PICARD<br>1 hour (range: 0.75-1.25) 1/20 nodes   | 0.00034                    | 0.00026-0.00043 | TCAG Bioinformatics manager, 2018 | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |
| Recalibration- GATK (step 1)<br>1.5 hours (range: 4-6) 1/20 nodes  | 0.00051                    | 0.00043-0.00060 | TCAG Bioinformatics manager, 2018 | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |
| Recalibration – GATK (step 2)<br>0.5 hour (range: 0.25-0.75) 2 nodes   | 0.00685                    | 0.00342-0.0103  | TCAG Bioinformatics manager, 2018 | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |

| Cost Items  | Quantity of Use per Sample |                    |   | Price (CAD \$/unit) |              |                                       |
|---|----------------------------|--------------------|---|---------------------|--------------|---------------------------------------|
|   | Estimate                   | Range              | Source                                    | Estimate            | Range        | Source                                |
| Post-recalibration merge – GATK<br>0.5 hour (range: 0.25-0.75) 1/20 nodes   | 0.000171                   | 0.0000856-0.000257 | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| Indel Realignment (GATK)<br>0.5 hour (range: 0.25-0.75) 2 nodes   | 0.01                       | 0.00342-0.0103     | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| SNV/indel variant calling - (GATK haplotype caller)<br>0.33 hour (range: 0.3-0.37) 2 nodes  | 0.00456                    | 0.00411-0.00507    | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| Annotation (ANNOVAR)<br>0.5 hour (range: 0.25-0.75) 0.2 nodes   | 0.00069                    | 0.000342-0.00103   | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| <b>Filtering &amp; Triage (Units: minutes)</b>  |                            |                    |   |                     |              |                                       |
| Filtration of primary variants  | 55                         | Not assigned       | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20% | Public Sector Salary Disclosure, 2017 |
| Filtration of secondary variants  | 5                          | Not assigned       | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20% | Public Sector Salary Disclosure, 2017 |
| <b>Clinical Interpretation (Units: minutes)</b>   |                            |                    |   |                     |              |                                       |
| Clinical classification of primary variants<br>15 minutes if no variants are found, otherwise 30 minutes per variant. On average, 2 variants are found (range: 0-4) | 60                         | 15-120             | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20% | Public Sector Salary Disclosure, 2017 |
| Clinical classification of secondary variants<br>30 minutes per case (range: 20-40), found in 4% of cases (range: 3-5%)   | 1.2                        | 0.6-2.0            | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20% | Public Sector Salary Disclosure, 2017 |
| <b>Report Writing (Units: minutes)</b>  |                            |                    |   |                     |              |                                       |
| Addressing primary variants   | 45                         | 15-75              | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20% | Public Sector Salary Disclosure, 2017 |

| Cost Items  | Quantity of Use per Sample |                |   | Price (CAD \$/unit) |               |   |
|---|----------------------------|----------------|---|---------------------|---------------|---|
|   | Estimate                   | Range          | Source                                    | Estimate            | Range         | Source  |
| 15 minutes base plus 15 minutes per variant. On average, 2 variants are found (range: 0-4)              |                            |                |   |                     |               |   |
| Addressing secondary variants<br>30 minutes per case (range: 20-40), found in 4% of cases (range: 3-5%) | 1.2                        | 0.6-2.0        | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20%  | Public Sector Salary Disclosure, 2017           |
| <b>Large Equipment</b>  |                            |                |   |                     |               |   |
| Illumina NextSeq® 550   | 1/all tests                | Not applicable | TCAG lab manager                          | 201250              | 181125-221375 | TCAG lab manager, 2018                          |
| 1-year service contract   | 1/all tests                | Not applicable | TCAG lab manager                          | 17940               | 16146-19734   | TCAG lab manager, 2018                          |
| Agilent BioAnalyzer/Tape station  | 1/all tests                | Not applicable | Agilent representative                    | 38500               | 34000-43000   | Agilent representative/TCAG lab manager, 2015   |
| <b>Small Equipment</b>  |                            |                |   |                     |               |   |
| Tube microcentrifuge  | 1/all tests                | Not applicable | TCAG lab manager                          | 2250                | 2000-2500     | TCAG lab manager, 2014                          |
| Plate microcentrifuge   | 1/all tests                | Not applicable | TCAG lab manager                          | 5000                | 4500-5500     | TCAG lab manager, 2014                          |
| Thermomixer   | 1/all tests                | Not applicable | TCAG lab manager                          | 5000                | 4500-5500     | TCAG lab manager, 2014                          |
| Vortex  | 1/all tests                | Not applicable | TCAG lab manager                          | 450                 | 400-500       | TCAG lab manager, 2014                          |
| Pipette sets  | 2/all tests                | Not applicable | TCAG lab manager                          | 1600                | 1400-2000     | TCAG lab manager, 2014                          |
| Magnet particle concentrator for tubes  | 1/all tests                | Not applicable | TCAG lab manager                          | 700                 | 630-770       | TCAG lab manager, 2014                          |
| Thermocyclers   | 2/all tests                | Not applicable | TCAG lab manager                          | 3000                | 2700-3300     | TCAG lab manager, 2014                          |
| <b>Supplies</b>   |                            |                |   |                     |               |   |
| Shipping & Handling   | 1                          | Not assigned   | TCAG lab manager                          | 37.61               | 33.80-41.40   | FedEx, 2018                                     |
| SureSelect Baits (96 reactions)   | 1                          | Not assigned   | TCAG lab manager                          | 195                 | 175.50-214.50 | TCAG lab manager, 2018                          |
| SureSelect Library prep (96 reactions)  | 1                          | Not assigned   | TCAG lab manager                          | 22.50               | 20.30-24.80   | TCAG lab manager, 2018                          |
| Other library prep consumables  | 1                          | Not assigned   | TCAG lab manager                          | 70.0                | 63.00-77.00   | TCAG lab manager, 2018                          |
| Sequencing reagents (8 samples per lane)  | 1                          | Not assigned   | TCAG lab manager                          | 707.83              | 637.00-778.60 | TCAG lab manager 2018                           |
| <b>Follow-up (proportion of cases)</b>  |                            |                |   |                     |               |   |
| Sanger sequencing (two tests per person, proband and two parents)                                       | 0.5                        | Not assigned   | DPLM Associate director, Genomic Analysis | 53.33               | 48.00-58.70   | DPLM Associate director, Genomic Analysis, 2018 |

| Cost Items   | Quantity of Use per Sample |            |                             | Price (CAD \$/unit) |             |                                   |
|--|----------------------------|------------|-----------------------------|---------------------|-------------|-----------------------------------|
|  | Estimate                   | Range      | Source                      | Estimate            | Range       | Source                            |
| <b>Bioinformatics</b>  |                            |            |                             |                     |             |                                   |
| <b>Bioinformatics File Storage (Units: GB per year)</b>  |                            |            |                             |                     |             |                                   |
| Notes: 1. Resource use volume was calculated by multiplying the file size by the storage time (in years).  |                            |            |                             |                     |             |                                   |
| 2. Price was given in dollars (\$) per GB per year.  |                            |            |                             |                     |             |                                   |
| trimmed fastq<br>file size: 9 GB; storage time: 9 months (range: 6-12)   | 6.75                       | 4.5-9.0    | TCAG Bioinformatics manager | 0.4                 | 0.36-0.44   | TCAG Bioinformatics manager, 2014 |
| final rem-dup, recalibrated, locally re-aligned BAM file<br>file size: 9 GB; storage time: 9 months (range: 6-12)  | 4.5                        | 3.0-6.0    | TCAG Bioinformatics manager | 0.4                 | 0.36-0.44   | TCAG Bioinformatics manager, 2014 |
| <b>Bioinformatics Computation Use (Units: CPU time per hour)</b>   |                            |            |                             |                     |             |                                   |
| Notes: 1. Resource use volume was calculated by multiplying the number of jobs by the number of cores per job and time (hours) required to complete a job. 25% (range 0 to 50%) was added to account for additional processing time to avoid operating at full capacity.   |                            |            |                             |                     |             |                                   |
| 2. Scalar Decisions, Inc. quote of \$26,804 per node (20 cores) or \$1340.20 per core (including warranty) was used. A straight line depreciation method over 5 years was applied and the price per CPU per year was calculated to be \$5360.80. This price was converted to price per CPU per hour by dividing \$5360.80 by (365*24). |                            |            |                             |                     |             |                                   |
| Alignment<br>1 job, 54 GB RAM per job, 19 core per job, 0.5 hour to complete, >>1 nodes required   | 11.875                     | 9.5-14.25  | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | Scalar quote, 2016                |
| Mark Duplicates – PICARD<br>1 job, 5 GB RAM per job, 2 cores per job, 1 hour to complete, >> 1/20 node required  | 2.5                        | 2-3        | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | Scalar quote, 2016                |
| Recalibration- GATK (step 1)<br>1 job, 30 GB RAM per job, 1 core per job, 1.5 hours to complete, >>1/20 node required  | 1.875                      | 1.5-2.25   | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | Scalar quote, 2016                |
| Recalibration – GATK (step 2)  | 14.375                     | 11.5-17.25 | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | Scalar quote, 2016                |

| Cost Items  | Quantity of Use per Sample |           |                             | Price (CAD \$/unit) |             |                    |
|---|----------------------------|-----------|-----------------------------|---------------------|-------------|--------------------|
|   | Estimate                   | Range     | Source                      | Estimate            | Range       | Source             |
| 23 jobs, 30 GB RAM per job, 1 core per job, 0.5 hour to complete, >>2 node required   |                            |           |                             |                     |             |                    |
| Post-recalibration merge - (GATK)<br>1 job, 10 GB RAM per job, 2 core per job, 0.5 hour to complete, >>1/20 node required                 | 1.25                       | 1-1.5     | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | Scalar quote, 2016 |
| Indel Realignment (GATK)<br>23 jobs, 10 GB RAM per job, 2 core per job, 0.5 hour to complete, >>2 node required                           | 28.75                      | 23-34.5   | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | Scalar quote, 2016 |
| SNV/indel variant calling - GATK haplotype caller<br>23 jobs, 55 GB RAM per job, 1 core per job, 0.33 hour to complete, >>2 node required | 9.58                       | 7.67-11.5 | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | Scalar quote, 2016 |
| Annotation (ANNOVAR)<br>1 job, 32 GB RAM per job, 8 cores per job, 0.5 hour to complete, >>1/5 node required                              | 5                          | 4-6       | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | Scalar quote, 2016 |

Abbreviations: WES, Whole Exome Sequencing; TCAG, The Centre for Applied Genomics; DPLM, The Department of Paediatric Laboratory Medicine; FTE, Full-time equivalent; MIS, Management Information Systems; GATK, Genome Analysis Toolkit; SNV, Single nucleotide variant.

**Appendix 9. Resource use and unit prices for WGS-proband inputs, Illumina HiSeq X™**

| Cost Items  | Quantity of Use per Sample |              |                    | Price (CAD \$/unit) |              |  |
|---|----------------------------|--------------|--------------------|---------------------|--------------|--|
|   | Estimate                   | Range        | Source             | Estimate            | Range        | Source   |
| <b>Labour</b>   |                            |              |                    |                     |              |  |
| <b>Specimen Preparation (Units: minutes)</b>  |                            |              |                    |                     |              |  |
| 10120 Pediatric venipuncture  | 7.6                        | Not assigned | MIS Standards 2016 | Confidential        | -22%to +28%  | SickKids wage information, 2014; 1.5% annual increase to 2017/18 |
| 11090 Packaging with testing documentation  | 1.0                        | Not assigned | MIS Standards 2016 | Confidential        | -22% to +28% | SickKids wage information, 2014; 1.5% annual increase to 2017/18 |
| 11500 Service recipient primary registration  | 1.8                        | Not assigned | MIS Standards 2016 | Confidential        | -20% to +20% | SickKids wage information, 2018                                  |
| 10360 Printing and sorting of specimen labels (per label)   | 0.4                        | Not assigned | MIS Standards 2016 | Confidential        | -20% to +20% | SickKids wage information, 2018                                  |
| 88550 Creation of family/service recipient folder (add sample to existing family folder, assign a new number and retrieve or make a new folder) (once per folder) | 5.0                        | Not assigned | MIS Standards 2016 | Confidential        | -20% to +20% | SickKids wage information, 2018                                  |
| 11090 Packaging with testing documentation  | 1.0                        | Not assigned | MIS Standards 2016 | Confidential        | -20% to +20% | SickKids wage information, 2018                                  |
| 11500 Service recipient limited registration  | 1.8                        | Not assigned | MIS Standards 2016 | Confidential        | -20% to +20% | SickKids wage information, 2018                                  |
| <b>Library preparation (Units: minutes)</b>   |                            |              |                    |                     |              |  |
| Note: Total time per task was divided by the number of samples per batch. The number of samples per batch was assumed to be 48.                                   |                            |              |                    |                     |              |  |
| DNA quantification (20 min total)   | 0.4                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| Pre-prep reagents (20 min total)  | 0.4                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| Shearing (20 min total)   | 0.4                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| Purification (40 min total)   | 0.8                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| End repair (40 min total)   | 0.8                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| A-tailing (40 min total)  | 0.8                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| Adapter ligation (45 min total)   | 0.9                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |



| Cost Items   | Quantity of Use per Sample |               |                                   | Price (CAD \$/unit) |              |                                   |
|--|----------------------------|---------------|-----------------------------------|---------------------|--------------|-----------------------------------|
|  | Estimate                   | Range         | Source                            | Estimate            | Range        | Source                            |
| <b>Sequencing (Units: minutes)</b>   |                            |               |                                   |                     |              |                                   |
| Note: Total time per task was divided by the number of samples per lane. The number of samples per lane was assumed to be 16.  |                            |               |                                   |                     |              |                                   |
| HiSeq wash (30 min total)  | 1.9                        | Not assigned  | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| Sequencing prep (30 min total)   | 1.9                        | Not assigned  | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| HiSeq post-run wash (45 min total)   | 2.8                        | Not assigned  | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| Run quality control (15 min total)   | 0.9                        | Not assigned  | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| cBot (30 min total)  | 1.9                        | Not assigned  | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| <b>Bioinformatics (Units: minutes)*</b>  |                            |               |                                   |                     |              |                                   |
| Note: resource use estimate is based on average output, 72 genomes per month. The lower bound was based on 96 genomes and the upper bound was based on 64 genomes.   |                            |               |                                   |                     |              |                                   |
| Variant calling (1 FTE hour for 64-96 genomes a month)   | 116.67                     | 87.54-131.25  | TCAG Bioinformatics manager       | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |
| Annotation (0.25 FTE hour for 64-96 genomes a month)   | 29.17                      | 21.875-32.813 | TCAG Bioinformatics manager       | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |
| <b>Bioinformatics Maintenance (Units: minutes)*</b>  |                            |               |                                   |                     |              |                                   |
| Note: One full time equivalent (FTE) hour to support one node per year was assumed. Resource use in minutes was calculated by multiplying the time to complete a job by the time needed to support the required number of nodes per job. For example, if a job required 3.2 hours and 1 node, then the resource use for the job is calculated as follows: 60 FTE minutes in an hour per year/ (365*24 hours in a year) *1 hour to support 1 node*3.2 hours (i.e. number of minutes per year to support a 192 minutes job on 1 node). The lower bound of resource use was based on 75% of the computation time and the upper bound was based on 125% of computation time. |                            |               |                                   |                     |              |                                   |
| Alignment<br>12 hours (range: 9.0-15.0) 1 node   | 0.0822                     | 0.06-0.10     | TCAG Bioinformatics manager, 2018 | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |
| Mark Duplicates -PICARD<br>5 hours (range: 3.75-6.25) 1/20 nodes   | 0.0017                     | 0.0013-0.0021 | TCAG Bioinformatics manager, 2018 | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |
| Recalibration (GATK)<br>2 hours (range: 1.5-2.5) 2 nodes   | 0.0274                     | 0.0205-0.0342 | TCAG Bioinformatics manager, 2018 | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |
| Post-recalibration merge - Genome Analysis Toolkit (GATK)<br>1 hour (range: 0.75-1.25) 1/20 nodes  | 0.0003                     | 0.0003-0.0004 | TCAG Bioinformatics manager, 2018 | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |

| Cost Items  | Quantity of Use per Sample |                   |   | Price (CAD \$/unit) |              |                                       |
|---|----------------------------|-------------------|---|---------------------|--------------|---------------------------------------|
|   | Estimate                   | Range             | Source                                    | Estimate            | Range        | Source                                |
| Indel Realignment (GATK)<br>1.5 hours (range: 1.13-1.875) 2 nodes                         | 0.0205                     | 0.0154-0.0257     | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| SNV/indel variant calling - (GATK haplotype caller)<br>3 hours (range: 2.25-3.75) 2 nodes | 0.0411                     | 0.0308-0.0514     | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| Annotation (ANNOVAR)<br>1.5 hours (range: 1.13-1.875) 1/5 nodes                           | 0.0021                     | 0.0015-0.0026     | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| CNV Detection (Custom)<br>15 hours (range: 11.25-18.75) 1/20 nodes                        | 0.0051                     | 0.0039-0.0064     | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| CNV Annotation (Custom)<br>1/12 hours (range: 0.0625-0.104) 1/20 nodes                    | 0.0000285                  | 0.000021-0.000036 | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| SV Detection (MANTA)<br>1 hour (range: 0.75-1.25) 1 node                                  | 0.0068                     | 0.0051-0.0086     | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| SV Annotation (Custom)<br>1/12 hours (range: 0.0625-0.104) 1/20 nodes                     | 0.0000285                  | 0.000021-0.000036 | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| <b>Filtering &amp; Triage (Units: minutes)</b>  |                            |                   |   |                     |              |                                       |
| Filtration of primary variants  | 55                         | Not assigned      | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20% | Public Sector Salary Disclosure, 2017 |
| Filtration of secondary variants  | 5                          | Not assigned      | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20% | Public Sector Salary Disclosure, 2017 |
| <b>Clinical Interpretation (Units: minutes)</b>   |                            |                   |   |                     |              |                                       |
| Clinical classification of primary variants   | 75                         | 15-135            | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20% | Public Sector Salary Disclosure, 2017 |

| Cost Items  | Quantity of Use per Sample |                |   | Price (CAD \$/unit) |                 |   |
|---|----------------------------|----------------|---|---------------------|-----------------|---|
|   | Estimate                   | Range          | Source                                    | Estimate            | Range           | Source  |
| 15 minutes base plus 30 minutes per variant. On average, 2 variants are found (range: 0-4)                                |                            |                |   |                     |                 |   |
| Clinical classification of secondary variants<br>30 minutes per case (range: 20-40), found in 4% of cases (range: 3-5%)   | 1.2                        | 0.6-2.0        | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20%    | Public Sector Salary Disclosure, 2017         |
| <b>Report Writing (Units: minutes)</b>  |                            |                |   |                     |                 |   |
| Addressing primary variants<br>15 minutes base plus 15 minutes per variant. On average, 2 variants are found (range: 0-4) | 45                         | 15-75          | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20%    | Public Sector Salary Disclosure, 2017         |
| Addressing secondary variants<br>30 minutes per case (range: 20-40), found in 4% of cases (range: 3-5%)                   | 1.2                        | 0.6-2.0        | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20%    | Public Sector Salary Disclosure, 2017         |
| <b>Large Equipment</b>  |                            |                |   |                     |                 |   |
| Illumina HiSeq X™   | 1/all tests                | Not applicable | TCAG Lab manager                          | 1150000             | 1035000-1265000 | TCAG Lab manager, 2015                        |
| 1-year service contract   | 1/all tests                | Not applicable | TCAG Lab manager                          | 119025              | 107123-130928   | TCAG Lab manager, 2015                        |
| Agilent BioAnalyzer/Tape station  | 1/all tests                | Not applicable | Agilent representative                    | 38500               | 34000-43000     | Agilent representative/TCAG Lab manager, 2015 |
| <b>Small Equipment</b>  |                            |                |   |                     |                 |   |
| Tube microcentrifuge  | 1/all tests                | Not applicable | TCAG lab manager                          | 2250                | 2000-2500       | TCAG lab manager, 2014                        |
| Plate microcentrifuge   | 1/all tests                | Not applicable | TCAG lab manager                          | 5000                | 4500-5500       | TCAG lab manager, 2014                        |
| Thermomixer   | 1/all tests                | Not applicable | TCAG lab manager                          | 5000                | 4500-5500       | TCAG lab manager, 2014                        |
| Vortex  | 1/all tests                | Not applicable | TCAG lab manager                          | 450                 | 400-500         | TCAG lab manager, 2014                        |
| Pipette sets  | 2/all tests                | Not applicable | TCAG lab manager                          | 1600                | 1400-2000       | TCAG lab manager, 2014                        |
| Magnet particle concentrator for tubes  | 1/all tests                | Not applicable | TCAG lab manager                          | 700                 | 630-770         | TCAG lab manager, 2014                        |
| Thermocyclers   | 2/all tests                | Not applicable | TCAG lab manager                          | 3000                | 2700-3300       | TCAG lab manager, 2014                        |
| <b>Supplies</b>   |                            |                |   |                     |                 |   |
| Shipping & handling   | 1                          | Not assigned   | TCAG lab manager                          | 37.61               | 33.80-41.40     | FedEx, 2018                                   |

| Cost Items  | Quantity of Use per Sample |              |   | Price (CAD \$/unit) |               |   |
|---|----------------------------|--------------|---|---------------------|---------------|---|
|   | Estimate                   | Range        | Source                                    | Estimate            | Range         | Source  |
| Illumina Nano DNA library prep  | 1                          | Not assigned | TCAG lab manager                          | 30.0                | 27.0-33.0     | TCAG lab manager, 2015                          |
| Other library prep consumables per sample   | 1                          | Not assigned | TCAG lab manager                          | 50.0                | 45.00-55.00   | TCAG lab manager, 2015                          |
| Sequencing reagents (1 sample per lane)   | 1                          | Not assigned | TCAG lab manager                          | 1290.0              | 1161.0-1419.0 | TCAG lab manager, 2015                          |
| <b>Follow-up (proportion of cases)</b>  |                            |              |   |                     |               |   |
| Note: Price for Sanger sequencing and qPCR test included three tests, proband and two parents, with two sequences per person  |                            |              |   |                     |               |   |
| Sanger sequencing (two tests per person, proband and two parents)   | 0.5                        | Not assigned | DPLM Associate director, Genomic Analysis | 53.33               | 48.00-58.67   | DPLM Associate director, Genomic Analysis, 2018 |
| qPCR follow-up (proband and two parents)  | 0.1                        | Not assigned | Team leader, Molecular Genetics           | 223.90              | 201.51-246.29 | Expert opinion, 2018                            |
| <b>Bioinformatics*</b>  |                            |              |   |                     |               |   |
| <b>Bioinformatics File Storage (Units: GB/year)</b>   |                            |              |   |                     |               |   |
| Notes: 1. Resource use volume was calculated by multiplying the file size by the storage time (in years).<br>2. Price was given in dollars (\$) per GB per year.  |                            |              |   |                     |               |   |
| trimmed fastq<br>file size: 120 GB; average storage time: 9 months (range: 6-12)  | 90                         | 60-120       | TCAG Bioinformatics analyst               | 0.4                 | 0.36-0.44     | TCAG Bioinformatics manager, 2014               |
| final rem-dup, recalibrated, locally re-aligned BAM file<br>file size: 80 GB; average storage time: 9 months (range: 6-12)  | 60                         | 40-80        | TCAG Bioinformatics analyst               | 0.4                 | 0.36-0.44     | TCAG Bioinformatics manager, 2014               |
| <b>Bioinformatics Computation Use (Units: CPU time per hour)</b>  |                            |              |   |                     |               |   |
| Notes: 1. Resource use volume was calculated by multiplying the number of jobs by the number of cores per job and time (hours) required to complete a job. 25% (range 0 to 50%) was added to account for additional processing time to avoid operating at full capacity.<br>2. The cost of each compute node to TCAG was \$26,804, including warranty. A straight line depreciation method over 5 years was applied and the price per CPU per year was calculated to be \$5,360.80. This price was converted to price per CPU per hour by dividing \$5360.80 by (365*24). |                            |              |   |                     |               |   |
| Alignment (BWA)<br>1 job, 54 GB RAM per job, 19 core per job, 12 hours to complete, 1 node required   | 285                        | 228-342      | TCAG Bioinformatics manager               | 0.612               | 0.551-0.673   | TCAG Bioinformatics analyst, 2016               |
| Mark Duplicates -PICARD   | 12.5                       | 10-15        | TCAG Bioinformatics manager               | 0.612               | 0.551-0.673   | TCAG Bioinformatics analyst, 2016               |

| Cost Items  | Quantity of Use per Sample |             |                             | Price (CAD \$/unit) |             |                                   |
|---|----------------------------|-------------|-----------------------------|---------------------|-------------|-----------------------------------|
|   | Estimate                   | Range       | Source                      | Estimate            | Range       | Source                            |
| 1 job, 5 GB RAM per job, 2 core per job, 5 hours to complete, 1/20 node required  |                            |             |                             |                     |             |                                   |
| Recalibration (GATK)<br>23 jobs, 30 GB RAM per job, 1 core per job, 2 hours to complete, 2 nodes required                                     | 57.5                       | 46-69       | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | TCAG Bioinformatics analyst, 2016 |
| Post-recalibration merge - Genome Analysis Toolkit (GATK)<br>1 job, 10 GB RAM per job, 2 core per job, 1 hour to complete, 1/20 node required | 2.5                        | 2.0-3.0     | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | TCAG Bioinformatics analyst, 2016 |
| Indel Realignment (GATK)<br>23 jobs, 10 GB RAM per job, 2 core per job, 1.5 hours to complete, 2 nodes required                               | 86.25                      | 69-103.5    | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | TCAG Bioinformatics analyst, 2016 |
| SNV/indel variant calling - (GATK haplotype caller)<br>23 jobs, 55 GB RAM per job, 1 core per job, 3 hours to complete, 2 nodes required      | 86.25                      | 69-103.5    | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | TCAG Bioinformatics analyst, 2016 |
| Annotation (ANNOVAR)<br>1 job, 32 GB RAM per job, 8 core per job, 1.5 hours to complete, 1/5 node required                                    | 15                         | 12-18       | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | TCAG Bioinformatics analyst, 2016 |
| CNV Detection (Custom)<br>2 jobs, 32 GB RAM per job, 1 core per job, 15 hours to complete, 1/20 node required                                 | 37.5                       | 30-45       | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | TCAG Bioinformatics analyst, 2016 |
| CNV Annotation (Custom)   | 0.104                      | 0.083-0.125 | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | TCAG Bioinformatics analyst, 2016 |

| Cost Items   | Quantity of Use per Sample |             |                             | Price (CAD \$/unit) |             |                                   |
|--|----------------------------|-------------|-----------------------------|---------------------|-------------|-----------------------------------|
|  | Estimate                   | Range       | Source                      | Estimate            | Range       | Source                            |
| 1 job, 16 GB RAM per job, 1 core per job, 1/12 hours to complete, 1/20 node required                         |                            |             |                             |                     |             |                                   |
| SV Detection (MANTA)<br>1 job, 40 GB RAM per job, 20 core per job, 1 hour to complete, 1 node required       | 25                         | 20-30       | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | TCAG Bioinformatics analyst, 2016 |
| SV Annotation (Custom)<br>1 job, 16GB RAM per job, 1 core per job, 1/12 hour to complete, 1/20 node required | 0.104                      | 0.083-0.125 | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | TCAG Bioinformatics analyst, 2016 |

Abbreviations: WGS, Whole Genome Sequencing; TCAG, The Centre for Applied Genomics; DPLM, The Department of Paediatric Laboratory Medicine; FTE, Full-time equivalent; qPCR, Real-time polymerase chain reaction; MIS, Management Information Systems; GATK, Genome Analysis Toolkit; SNV, Single nucleotide variant.

**Appendix 10. Resource use and unit prices for WGS-trio inputs, Illumina HiSeq X™**

| Cost Items  | Quantity of Use per Sample |              |                    | Price (CAD \$/unit) |              |  |
|---|----------------------------|--------------|--------------------|---------------------|--------------|--|
|   | Estimate                   | Range        | Source             | Estimate            | Range        | Source   |
| <b>Labour</b>   |                            |              |                    |                     |              |  |
| <b>Specimen Preparation (Units: minutes)</b>  |                            |              |                    |                     |              |  |
| 10120 Pediatric venipuncture  | 22.8                       | Not assigned | MIS Standards 2016 | Confidential        | -22%to +28%  | SickKids wage information, 2014; 1.5% annual increase to 2017/18 |
| 11090 Packaging with testing documentation  | 3                          | Not assigned | MIS Standards 2016 | Confidential        | -22% to +28% | SickKids wage information, 2014; 1.5% annual increase to 2017/18 |
| 11500 Service recipient primary registration  | 5.4                        | Not assigned | MIS Standards 2016 | Confidential        | -20% to +20% | SickKids wage information, 2018                                  |
| 10360 Printing and sorting of specimen labels (per label)   | 1.2                        | Not assigned | MIS Standards 2016 | Confidential        | -20% to +20% | SickKids wage information, 2018                                  |
| 88550 Creation of family/service recipient folder (add sample to existing family folder, assign a new number and retrieve or make a new folder) (once per folder) | 15                         | Not assigned | MIS Standards 2016 | Confidential        | -20% to +20% | SickKids wage information, 2018                                  |
| 11090 Packaging with testing documentation  | 3                          | Not assigned | MIS Standards 2016 | Confidential        | -20% to +20% | SickKids wage information, 2018                                  |
| 11500 Service recipient limited registration  | 5.4                        | Not assigned | MIS Standards 2016 | Confidential        | -20% to +20% | SickKids wage information, 2018                                  |
| <b>Library preparation (Units: minutes)</b>   |                            |              |                    |                     |              |  |
| Note: Total time per task was divided by the number of samples per batch. The number of samples per batch was assumed to be 48.                                   |                            |              |                    |                     |              |  |
| DNA quantification (20 min total)   | 1.3                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| Pre-prep reagents (20 min total)  | 1.3                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| Shearing (20 min total)   | 1.3                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| Purification (40 min total)   | 2.5                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| End repair (40 min total)   | 2.5                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| A-tailing (40 min total)  | 2.5                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |
| Adapter ligation (45 min total)   | 2.8                        | Not assigned | TCAG lab manager   | Confidential        | -10% to +10% | TCAG lab manager, 2018   |

| Cost Items  | Quantity of Use per Sample |                 |                                   | Price (CAD \$/unit) |              |                                   |
|---|----------------------------|-----------------|-----------------------------------|---------------------|--------------|-----------------------------------|
|   | Estimate                   | Range           | Source                            | Estimate            | Range        | Source                            |
| <b>Sequencing (Units: minutes)</b>  |                            |                 |                                   |                     |              |                                   |
| Note: Total time per task was divided by the number of samples per lane. The number of samples per lane was assumed to be 16.   |                            |                 |                                   |                     |              |                                   |
| HiSeq wash (30 min total)   | 5.6                        | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| Sequencing prep (30 min total)  | 5.6                        | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| HiSeq post-run wash (45 min total)  | 8.4                        | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| Run quality control (15 min total)  | 2.8                        | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| cBot (30 min total)   | 5.6                        | Not assigned    | TCAG lab manager                  | Confidential        | -10% to +10% | TCAG lab manager, 2018            |
| <b>Bioinformatics (Units: minutes)*</b>   |                            |                 |                                   |                     |              |                                   |
| Note: resource use estimate is based on average output, 72 genomes per month. The lower bound was based on 96 genomes and the upper bound was based on 64 genomes.  |                            |                 |                                   |                     |              |                                   |
| Variant calling (1 FTE hour for 64-96 genomes a month)  | 116.67                     | 87.5-131.25     | TCAG Bioinformatics manager       | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |
| Annotation (0.25 FTE hour for 64-96 genomes a month)  | 29.17                      | 21.875-32.813   | TCAG Bioinformatics manager       | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |
| <b>Bioinformatics Maintenance (Units: minutes)*</b>   |                            |                 |                                   |                     |              |                                   |
| Note: One full time equivalent (FTE) hour to support one node per year was assumed. Resource use in minutes was calculated by multiplying the time to complete a job by the time needed to support the required number of nodes per job. For example, if a job required 3.2 hours and 1 nodes, then the resource use for the job is calculated as follows: 60 FTE minutes in an hour per year/ (365*24 hours in a year)*1 hour to support 1 node*3.2 hours (i.e. number of minutes per year to support a 192 minute job on 1 node). The lower bound of resource use was based on 75% of the computation time and the upper bound was based on 125% of computation time. |                            |                 |                                   |                     |              |                                   |
| Alignment<br>12 hours (range: 9.0-15.0) 1 node  | 0.25                       | 0.18-0.31       | TCAG Bioinformatics manager, 2018 | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |
| Mark Duplicates –PICARD<br>5 hours (range: 3.75-6.25) 1/20 nodes  | 0.01                       | 0.004-0.01      | TCAG Bioinformatics manager, 2018 | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |
| Recalibration (GATK)<br>2 hours (range: 1.5-2.5) 2 nodes  | 0.08                       | 0.06-0.10       | TCAG Bioinformatics manager, 2018 | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |
| Post-recalibration merge - Genome Analysis Toolkit (GATK)<br>1 hour (range: 0.75-1.25) 1/20 nodes   | 0.0010                     | 0.00077-0.00128 | TCAG Bioinformatics manager, 2018 | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018 |



| Cost Items  | Quantity of Use per Sample |                    |   | Price (CAD \$/unit) |              |                                       |
|---|----------------------------|--------------------|---|---------------------|--------------|---------------------------------------|
|   | Estimate                   | Range              | Source                                    | Estimate            | Range        | Source                                |
| Indel Realignment (GATK)<br>1.5 hours (range: 1.13-1.875) 2 nodes   | 0.06                       | 0.05-0.08          | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| SNV/indel variant calling - (GATK haplotype caller)<br>3 hours (range: 2.25-3.75) 2 nodes   | 0.12                       | 0.09-0.15          | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| Annotation (ANNOVAR)<br>1.5 hours (range: 1.13-1.875) 1/5 nodes   | 0.01                       | 0.00462-0.0077     | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| CNV Detection (Custom)<br>15 hours (range: 11.25-18.75) 1/20 nodes  | 0.02                       | 0.012-0.019        | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| CNV Annotation (Custom)<br>1/12 hours (range: 0.0625-0.104) 1/20 nodes  | 0.0000856                  | 0.0000642-0.000107 | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| SV Detection (MANTA)<br>1 hour (range: 0.75-1.25) 1 node  | 0.02                       | 0.0154-0.0257      | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| SV Annotation (Custom)<br>1/12 hours (range: 0.0625-0.104) 1/20 nodes   | 0.0000856                  | 0.0000642-0.000107 | TCAG Bioinformatics manager, 2018         | Confidential        | -20% to +20% | TCAG Bioinformatics manager, 2018     |
| <b>Filtering &amp; Triage (Units: minutes)</b>  |                            |                    |   |                     |              |                                       |
| Filtration of primary variants  | 25                         | Not assigned       | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20% | Public Sector Salary Disclosure, 2017 |
| Filtration of secondary variants  | 5                          | Not assigned       | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20% | Public Sector Salary Disclosure, 2017 |
| <b>Clinical Interpretation (Units: minutes)</b>   |                            |                    |   |                     |              |                                       |
| Clinical classification of primary variants<br>15 minutes base plus 30 minutes per variant. On average, 2 variants are found (range: 0-4) | 75                         | 15-135             | DPLM Associate director, Genomic Analysis | Confidential        | -20% to +20% | Public Sector Salary Disclosure, 2017 |

| Cost Items  | Quantity of Use per Sample |                |  | Price (CAD \$/unit) |                     |   |
|---|----------------------------|----------------|--|---------------------|---------------------|---|
|   | Estimate                   | Range          | Source                                       | Estimate            | Range               | Source  |
| Clinical classification of secondary variants<br>30 minutes per case (range: 20-40),<br>found in 4% of cases (range: 3-5%)      | 1.2                        | 0.6-2.0        | DPLM Associate director,<br>Genomic Analysis | Confidential        | -20% to +20%        | Public Sector Salary<br>Disclosure, 2017          |
| <b>Report Writing (Units: minutes)</b>  |                            |                |  |                     |                     |   |
| Addressing primary variants<br>15 minutes base plus 15 minutes<br>per variant. On average, 2 variants<br>are found (range: 0-4) | 45                         | 15-75          | DPLM Associate director,<br>Genomic Analysis | Confidential        | -20% to +20%        | Public Sector Salary<br>Disclosure, 2017          |
| Addressing secondary variants<br>30 minutes per case (range: 20-40),<br>found in 4% of cases (range: 3-5%)                      | 1.2                        | 0.6-2.0        | DPLM Associate director,<br>Genomic Analysis | Confidential        | -20% to +20%        | Public Sector Salary<br>Disclosure, 2017          |
| <b>Large Equipment</b>  |                            |                |  |                     |                     |   |
| Illumina HiSeq X™   | 1/all tests                | Not applicable | TCAG Lab manager                             | 1150000             | 1035000-<br>1265000 | TCAG Lab manager, 2015                            |
| 1-year service contract   | 1/all tests                | Not applicable | TCAG Lab manager                             | 119025              | 107123-<br>130928   | TCAG Lab manager, 2015                            |
| Agilent BioAnalyzer/Tape station  | 1/all tests                | Not applicable | Agilent representative                       | 38500               | 34000-43000         | Agilent representative/<br>TCAG Lab manager, 2015 |
| <b>Small Equipment</b>  |                            |                |  |                     |                     |   |
| Tube microcentrifuge  | 1/all tests                | Not applicable | TCAG lab manager                             | 2250                | 2000-2500           | TCAG lab manager, 2014                            |
| Plate microcentrifuge   | 1/all tests                | Not applicable | TCAG lab manager                             | 5000                | 4500-5500           | TCAG lab manager, 2014                            |
| Thermomixer   | 1/all tests                | Not applicable | TCAG lab manager                             | 5000                | 4500-5500           | TCAG lab manager, 2014                            |
| Vortex  | 1/all tests                | Not applicable | TCAG lab manager                             | 450                 | 400-500             | TCAG lab manager, 2014                            |
| Pipette sets  | 2/all tests                | Not applicable | TCAG lab manager                             | 1600                | 1400-2000           | TCAG lab manager, 2014                            |
| Magnet particle concentrator for<br>tubes   | 1/all tests                | Not applicable | TCAG lab manager                             | 700                 | 630-770             | TCAG lab manager, 2014                            |
| Thermocyclers   | 2/all tests                | Not applicable | TCAG lab manager                             | 3000                | 2700-3300           | TCAG lab manager, 2014                            |
| <b>Supplies</b>   |                            |                |  |                     |                     |   |
| Shipping & Handling   | 3                          | Not assigned   | TCAG lab manager                             | 37.61               | 33.80-41.40         | FedEx, 2018                                       |
| Illumina Nano DNA library prep  | 3                          | Not assigned   | TCAG lab manager                             | 30.0                | 27.0-33.0           | TCAG lab manager, 2015                            |
| Other library prep consumables<br>per sample  | 3                          | Not assigned   | TCAG lab manager                             | 50.0                | 45.00-55.00         | TCAG lab manager, 2015                            |

| Cost Items  | Quantity of Use per Sample |              |   | Price (CAD \$/unit) |               |   |
|---|----------------------------|--------------|---|---------------------|---------------|---|
|   | Estimate                   | Range        | Source                                    | Estimate            | Range         | Source  |
| Sequencing reagents (1 sample per lane)   | 3                          | Not assigned | TCAG lab manager                          | 1290.0              | 1161.0-1419.0 | TCAG lab manager, 2015                          |
| <b>Follow-up (proportion of cases)</b>  |                            |              |   |                     |               |   |
| Note: Price for Sanger sequencing and qPCR test included three tests, proband and two parents, with two sequences per person  |                            |              |   |                     |               |   |
| Sanger sequencing (two tests per person, proband and two parents)   | 0.1                        | Not assigned | DPLM Associate director, Genomic Analysis | 53.33               | 48.00-58.67   | DPLM Associate director, Genomic Analysis, 2018 |
| qPCR followup (proband and two parents)   | 0.3                        | Not assigned | Team Leader, Molecular Genetics           | 223.90              | 201.51-246.29 | Expert opinion, 2018                            |
| <b>Bioinformatics*</b>  |                            |              |   |                     |               |   |
| <b>Bioinformatics File Storage (Units: GB/year)</b>   |                            |              |   |                     |               |   |
| Notes: 1. Resource use volume was calculated by multiplying the file size by the storage time (in years).<br>2. Price was given in dollars (\$) per GB per year.  |                            |              |   |                     |               |   |
| trimmed fastq<br>file size: 120 GB; average storage time: 9 months (range: 6-12)  | 270                        | 180-360      | TCAG Bioinformatics analyst               | 0.4                 | 0.36-0.44     | TCAG Bioinformatics manager, 2014               |
| final rem-dup, recalibrated, locally re-aligned BAM file<br>file size: 80 GB; average storage time: 9 months (range: 6-12)  | 180                        | 120-240      | TCAG Bioinformatics analyst               | 0.4                 | 0.36-0.44     | TCAG Bioinformatics manager, 2014               |
| <b>Bioinformatics Computation Use (Units: CPU time per hour)</b>  |                            |              |   |                     |               |   |
| Notes: 1. Resource use volume was calculated by multiplying the number of jobs by the number of cores per job and time (hours) required to complete a job. 25% (range 0 to 50%) was added to account for additional processing time to avoid operating at full capacity.<br>2. The cost of each compute node to TCAG was \$26,804, including warranty. A straight line depreciation method over 5 years was applied and the price per CPU per year was calculated to be \$5,360.80. This price was converted to price per CPU per hour by dividing \$5360.80 by (365*24). |                            |              |   |                     |               |   |
| Alignment (BWA)<br>1 job, 54 GB RAM per job, 19 core per job, 12 hours to complete, 1 node required   | 855                        | 684-1026     | TCAG Bioinformatics manager               | 0.612               | 0.551-0.673   | TCAG Bioinformatics analyst, 2016               |
| Mark Duplicates –PICARD<br>1 job, 5 GB RAM per job, 2 core per job, 5 hours to complete, 1/20 node required   | 37.5                       | 30-45        | TCAG Bioinformatics manager               | 0.612               | 0.551-0.673   | TCAG Bioinformatics analyst, 2016               |

| Cost Items   | Quantity of Use per Sample |            |                             | Price (CAD \$/unit) |             |                                   |
|--|----------------------------|------------|-----------------------------|---------------------|-------------|-----------------------------------|
|  | Estimate                   | Range      | Source                      | Estimate            | Range       | Source                            |
| Recalibration (GATK)<br>23 jobs, 30 GB RAM per job, 1 core per job, 2 hours to complete, 2 nodes required                                    | 172.5                      | 138-207    | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | TCAG Bioinformatics analyst, 2016 |
| Post-recalibration merge - Genome Analysis Toolkit (GATK)<br>1 job, 10 GB RAM per job, 2 core per job, 1hour to complete, 1/20 node required | 7.5                        | 6-9        | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | TCAG Bioinformatics analyst, 2016 |
| Indel Realignment (GATK)<br>23 jobs, 10 GB RAM per job, 2 core per job, 1.5 hours to complete, 2 nodes required                              | 258.75                     | 207-310.5  | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | TCAG Bioinformatics analyst, 2016 |
| SNV/indel variant calling - (GATK haplotype caller)<br>23 jobs, 55 GB RAM per job, 1 core per job, 3 hours to complete, 2 nodes required     | 258.75                     | 207-310.5  | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | TCAG Bioinformatics analyst, 2016 |
| Annotation (ANNOVAR)<br>1 job, 32 GB RAM per job, 8 core per job, 1.5 hours to complete, 1/5 node required                                   | 45                         | 36-54      | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | TCAG Bioinformatics analyst, 2016 |
| CNV Detection (Custom)<br>2 jobs, 32 GB RAM per job, 1 core per job, 15 hours to complete, 1/20 node required                                | 112.5                      | 90-135     | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | TCAG Bioinformatics analyst, 2016 |
| CNV Annotation (Custom)<br>1 job, 16 GB RAM per job, 1 core per job, 1/12 hour to complete, 1/20 node required                               | 0.3125                     | 0.25-0.375 | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | TCAG Bioinformatics analyst, 2016 |
| SV Detection (MANTA)   | 75                         | 60-90      | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | TCAG Bioinformatics analyst, 2016 |

| Cost Items   | Quantity of Use per Sample |            |                             | Price (CAD \$/unit) |             |                                   |
|--|----------------------------|------------|-----------------------------|---------------------|-------------|-----------------------------------|
|  | Estimate                   | Range      | Source                      | Estimate            | Range       | Source                            |
| 1 job, 40 GB RAM per job, 20 core per job, 1 hour to complete, 1 node required                               |                            |            |                             |                     |             |                                   |
| SV Annotation (Custom)<br>1 job, 16GB RAM per job, 1 core per job, 1/12 hour to complete, 1/20 node required | 0.3125                     | 0.25-0.375 | TCAG Bioinformatics manager | 0.612               | 0.551-0.673 | TCAG Bioinformatics analyst, 2016 |

Abbreviations: WGS, Whole Genome Sequencing; TCAG, The Centre for Applied Genomics; DPLM, The Department of Paediatric Laboratory Medicine; FTE, Full-time equivalent; qPCR, Real-time polymerase chain reaction; MIS, Management Information Systems; GATK, Genome Analysis Toolkit; SNV, Single nucleotide variant.