MOLECULAR HEALTH GUIDE | MOLECULAR HEALTH MENDEL | MOLECULAR HEALTH BRCA

TECH SHEET

Variant Detection Pipelines

MH Guide, MH Mendel, and MH BRCA provide optional Variant Detection Pipelines (VDP) to identify genetic variants and biomarkers with high precision and sensitivity in FASTQ and BAM files for DNAseq and RNAseq assays.

The **MH Guide VDP** is optimized for the identification of somatic variants from cancer samples using NGS panels. For whole exome sequencing (WES), a control sample is required and will also be analyzed. An RNAseq pipeline is available for higher-quality fusion calling.

The **MH Mendel VDP** and **MH BRCA VDP** are optimized for identification of germline variants from genomic samples using NGS panels, or using a whole exome sequencing (WES) approach.

Customers with their own VDPs, from the instrument or from other vendors, can upload VCF data in MH VCF format directly for analysis with MH Guide, MH Mendel, or MH BRCA.

Fast, scalable, and reliable variant detection

The MH Guide, MH Mendel, and MH BRCA VDPs are offered as a Software as a service (SaaS) in a CAP/CLIA-compliant environment. The hardware and software are optimized for massively parallel processing (MPP).

Major features

- Data can be uploaded via the MH Order portal, a user-friendly uploading interface, or using the MH SFTP process, optimized for high-throughput processing.
- Read alignment is based on standard (HG19, GRCh37), or proprietary GRCh37-based, or population-specific reference genomes (PHREGs), based on the 1000 genomes project.
- Variant filtering can be configured using a BED file specifying the target regions for SNV and indel calling, or using a gene filter list, or by filtering out non-coding variants that have no potential clinical significance.
- Detailed pipeline QC information is measured and shown on the user interface of MH Guide, MH Mendel, or MH BRCA. Additional information is available in log files.
- Analysis turnaround time (TOT) depends on the customer's network bandwidth, as well as on the target region size and coverage of the NGS experiment. As an indication of the TOT,
  - a typical unpaired gene panel can be processed in under 15 minutes.
  - an experiment with a more extensive target region, such as a WES analysis covering 20,000 genes, typically takes less than three hours.

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The MH Guide variant detection pipelines return an MH VCF format file. In combination with MH Guide, MH Mendel, or MH BRCA you receive a variant report in PDF format, or in JSON or XML format for further analysis or incorporation into existing processes.

Performance specifications

The performance and limits of detection were calculated using validation cases with a defined average sequencing coverage (somatic panel: 500x; somatic WES: 200x; germline: 100x). A similar performance can be expected for data that fulfill MH sequencing guideline requirements.

Analysis type	Variant type	Precision	Sensitivity	Validation based on
MH Mendel, MH BRCA, MH Guide VDP Germline	SNV	≥ 99%	≥ 99%	VAF ≥10%, 40x coverage at variant position (VP)
	Indel	≥ 99%	≥ 99%	VAF ≥10%, 40x coverage at VP
MH Guide VDP Somatic, Panel	SNV	≥ 99%	≥ 99%	Tumor content (TC) ≥20%, VAF ≥10%, 100x coverage at VP
	Indel	≥ 99%	≥ 99%	TC ≥20%, VAF ≥10%, 100x coverage at VP
	Fusion	≥ 99%	≥ 99%	TC ≥20%, supporting read pairs ≥5. Detection of selected, white-listed genomic "Fusion" variants
MH Guide VDP Somatic, WES	SNV	≥ 99%	≥ 98%	TC ≥20%, VAF ≥.10%, 100x coverage at VP
	Indel	≥ 99%	≥ 97%	TC ≥20%, VAF ≥10%, 100x coverage at VP
	CNA loss*	96%	96%	> 50% C. Detection of white-listed genomic "CNA loss" variants
	CNA gain*	99%	76%	>50% C. Detection of white-listed genomic "CNA gain" variants with copy number ≥4
	MSI-H*	100%	82%	Samples without sequencing QC warnings
MH Guide VDP Somatic, RNASeq	Fusion	≥ 95%	≥ 73%	Supporting reads ≥5. In general, recommended for "Fusion" detection in preference to DNAseq.

* MH recommends that CNA and MSI biomarkers should be orthogonally validated.

Remark: Information from additional test results can be added manually to the list of detected variants for analysis by MH Guide and MH Mendel, including SNVs, Indels, fusions, CNAs, protein expression, gene expression, methylation, wild types, tumor mutational burden, and microsatellite instability.