Oncomine

TEST DETAILS

The OnCORseq Cancer Gene Mutation Panel v2 assay is a targeted test for the identification of clinically significant somatic mutations including single nucleotide variants (SNVs), insertions and deletions (Indels), gene fusions and copy number alterations (CNAs) from DNA and RNA of 143 cancer-related genes (>2,500 amplicons) in solid tumors, using targeted next generation sequencing (NGS). The genes interrogated in the assay are categorized by genomic alterations into 73 hot spot genes (with SNVs and Indels), including 49 genes with focal CNAs, such as ERBB2, and 23 genes with known fusions such as ALK and ROS1 in NSCLC. The test also includes four pairs of 5’, 3’ expression imbalance assays for ALK, ROS1, RET and NTRK1. In addition, the test provides full coverage of 26 tumor suppressor genes. The OnCORseq assay was developed and distributed by Thermo Fisher as The Oncomine™ Comprehensive Assay (https://tools.thermofisher.com/content/sfs/brochures/oncomine-comprehensive-assay-flyer.pdf), and is aligned with the NCI-Molecular Analysis for Therapy Choice (NCI-MATCH) trial.

The panel facilitates the detection of actionable and targetable mutations in numerous cancer including lung, colorectal, thyroid, brain and skin cancer (melanoma) for appropriate diagnosis, prognosis and selection of therapy. The panel will also identify relevant genes that may have implications for enrollment of the patient in clinical trials.

The genes interrogated in the panel are listed below:
Specimen Requirements:

**Specimen Type: Paraffin embedded tissue sections**

- Tissue should be fixed in formalin and not exposed to decalcification solution. The paraffin block should contain at least a 3 mm area of tumor.
- The minimum amount of neoplastic cellularity acceptable in the tissue section is 10%.
- One H&E and 20 unstained sections are required for most of the tests. Please call the lab if you have questions.
- Slides should be properly labeled with a block label that matches the surgical pathology specimen number on the surgical pathology report.

**Specimen Type: Fixed Fine Needle Aspiration (FNA) samples**

- One H&E and 20 unstained sections from cell block are required.
- Fresh specimens should be collected into preservative solution.
- Slides should be properly labeled with a number that matches the specimen number on the cytology report.

*Note:* A surgical pathology and/or cytology report and completed requisition form must accompany all specimens.
**Ordering the Test**

Testing must be ordered by the Oncologist. Oncologists can request this test (*OnCORseq w/Interp Targeted Next Generation Sequencing*) through Surgical Pathology at the time the specimen is sent for diagnosis. Tumor specimens deemed most suitable for NGS testing by a pathologist will be send to the Genomic laboratory for further processing, nucleic acid extraction and testing.

**Test details and methodology**

Tumor DNA is enriched by extraction from unstained macro-dissected, formalin-fixed, paraffin-embedded (FFPE) tissue or cell sections of the patient’s specimen. An H&E stained section is used for neoplastic content evaluation and as a templet for macro-dissection. Specimens with a minimum of 20% neoplastic content in the area selected for macro-dissection as indicated by a pathologist are preferred for analysis. Minimum accepted neoplastic content is 10%.

Genomic DNA and RNA are then amplified, enriched and sequenced using next generation bidirectional sequencing technology on the S5 instrument (Thermo Fisher). The OnCORseq Cancer Panel hotspot v2 targeted gene panel is designed to detect mutations in >2,500 amplicons from 143 key cancer-related genes, including the detection of copy number gains and fusion driver genes. Quality control measures are implemented at multiple levels of the sequencing process. Annotation of variants is performed using the Ion Reporter Software v 5.0 (Thermo Fisher) and/or NextGene Software (Soft Genetics).

Genetic variants are then classified into three tiers based on their clinical relevance, using publicly available data and our own developed precision medicine knowledge base (PMKB). Mutation results are then reviewed and interpreted in the context of the clinical and pathological information by a board-certified molecular pathologist, who also releases the results (sign-out) in the laboratory information management system.

**Note:** This assay cannot unequivocally differentiate between somatic and constitutional (germline) mutations. Constitutional mutations identified incidentally will be reported separately according to ACMG guidelines for reporting incidental findings in clinical NGS testing ([https://www.acmg.net/docs/if_statement_final_7.24.13.pdf](https://www.acmg.net/docs/if_statement_final_7.24.13.pdf)).
Limitations of the assay

1. The OnCORseq mutation panel is designed to detect mutations in targeted regions/genes only as detailed in the table above. Pathogenic mutations outside the targeted regions/genes will not be detected.
2. The analytical sensitivity of the assay is approximately 5% (with a minimum neoplastic content of 10%); therefore, mutations present in a lower percentage of tumor cells may not be identified by this assay. Use of insufficient DNA or RNA template can result in low PCR product yields, and sequence signals may fall below detection limits.
3. The limit of detection is 5% variant allele frequency (VAF) at 400X coverage and 3% VAF at 1,000X coverage.
4. The ability of this assay to identify copy number alterations (CNAs) is reduced in cases with low tumor percentage (e.g., less than 50% tumor); in such cases, copy number alteration data (including the apparent absence of copy number alterations) should be interpreted with caution since the findings may not be representative of the tumor.
5. The ability of this assay to identify gene fusion alterations is reduced in specimens with low quality RNA. In such cases, negative findings should be interpreted with caution since the findings may not be representative of the tumor.
6. Selected genetic variants may require confirmation by an orthogonal method prior to reporting, which may cause a delay in reporting of results.
7. This assay cannot unequivocally differentiate between somatic and constitutional (germline) mutations. Constitutional mutations identified incidentally will be reported separately according to ACMG guidelines for reporting incidental findings in clinical NGS testing (https://www.acmg.net/docs/if_statement_final_7.24.13.pdf).

Disclaimer

The OnCORseq test was developed and its performance characteristics determined by the Englander Institute for Precision Medicine/NewYork Presbyterian Hospital Laboratories and approved by the New York-State Department of Health (NYS-DOH). Variants that are documented in the Catalogue of Somatic Mutations in Cancer (COSMIC) database in addition to non-COSMIC indel mutations in tumor suppressor genes are reported. Mutation details and corresponding COSMIC IDs for the genes in the panel can be obtained at http://cancer.sanger.ac.uk/cancergenome/projects/cosmic/ . The mutation nomenclature used for reporting of variants is based on the convention recommended by the Human Genome Variation Society (http://www.hgvs.org/mutnomen/). Mutations interpretations are drawn from a knowledgebase (https://pmkb.weill.cornell.edu) designed to collect and maintain interpretations of cancer gene mutations and facilitate reporting of NGS testing results.
**Turn Around Time:** 7 to 14 days

**Contact information:**

For further information on this assay or for questions on the results of the OnCORseq test please contact:

Jeffrey Catalano, Operations Manager ([Jpc2004@med.cornell.edu](mailto:Jpc2004@med.cornell.edu)), Phone: 646-962-3815

Dr. Wei Song, Assistant Director ([sow2005@med.cornell.edu](mailto:sow2005@med.cornell.edu)), Phone:

Dr. Hanna Rennert, Assistant Director ([har2006@med.cornell.edu](mailto:har2006@med.cornell.edu)), Phone: 212-746-6412