

#### CleanPlex® Ready-to-Use NGS Panels | Product Sheet

## OmniFusion<sup>TM</sup> RNA Lung Cancer Panel

# Fast and reliable assay for detecting known and novel gene fusions

#### Highlights

- Identification of known and novel RNA fusions associated with lung cancer Interrogate >530 known RNA fusions and additional novel RNA fusions using a single-primer method.
- Fast, streamlined workflow
   Generate sequencing-ready libraries in just 6.5 hours using a rapid, four-step protocol from extracted RNA to sequence ready libraries.
- Excellent performance with extremely low rRNA rate Prepare high-quality targeted NGS libraries using OmniFusion<sup>TM</sup> RNA Detection Technology to achieve minimal rRNA rate and over 90% on-target rate.

OmniFusion<sup>TM</sup> RNA Lung Cancer Panel for next generation sequencing enables detection of hundreds of known and unknown fusions associated with non-small cell lung cancer. In contrast to the AccuFusion<sup>TM</sup> panels, OmniFusion<sup>TM</sup> workflow allows detection of novel fusion partners. Compared to other methods such as qPCR, FISH, or Sanger sequencing, Paragon Genomics' targeted fusion sequencing method allows robust multiplexed detection of variants using minimum sample input and a simple workflow.

The OmniFusion<sup>TM</sup> single-primer amplification technology uses template switching technology to add an universal sequence to the 5' ends of RNA fragments. Combined with expertly designed targeted 3' primers specific to acceptor genes, the process eliminates the constraint of template length, and allows higher sensitivity even with damaged RNA fragments, such as FFPE RNA. The single sided amplification method enables the detection of any mutations on the RNA fragments, including novel ones.

#### OmniFusion<sup>TM</sup> RNA Lung Cancer Panel Specifications

| Parameter               | Specification   |
|-------------------------|---|
| Enrichment Method       | Multiplex PCR with single target specific primer  |
| Sequencing Platforms    | Illumina <sup>®</sup>   |
| Number of Fusion Genes  | 11 cancer driver genes ( ALK, CIT,<br>MBIP, MET, NRG1, NTRK1, NTRK3,<br>PDGFRA, RET, ROS1, TACC3)<br>3 control genes (B2M, GUSA, TBP) |
| Targets                 | >530 known fusions associated with NSCLC & novel fusions  |
| Variant Types           | Fusion down to 1% allele frequency  |
| Number of Amplicons     | 61  |
| Amplicon Size           | 200-700 bp (variable based on sample fragment size)   |
| Number of Primer Pools  | 1   |
| Input RNA Requirement   | 25-100 ng   |
| Sample Types            | FFPE, FNA, Fragmented RNA   |
| Total Assay Time        | 6.5 hours   |
| Hands-On Time           | 85 minutes  |
| rRNA rate               | < 1%  |
| On-Target Aligned Reads | ~93%  |

The OmniFusion<sup>TM</sup> technology was built upon the foundations of Paragon Genomics' CleanPlex® chemistry that produces high quality and clean libraries for sequencing. In addition, the OmniFusion<sup>TM</sup> technology sets itself apart from other NGS based methods by incorporating strategically optimized reverse transcription reagents to generate highly specific libraries with high on-target and mapping rates by minimizing rRNA rates. OmniFusion<sup>TM</sup> libraries allow highly-sensitive, efficient, and high-throughput fusion detection by sequencing.

#### OmniFusion Streamlined Workflow

The OmniFusion<sup>TM</sup> RNA Panels offer a simple and streamlined workflow. Starting from purified and quantitated RNA, the protocol starts with reverse transcription (RT) and template switching (TS), followed by the mPCR-based CleanPlex® workflow, which in total can be completed in less than 7 hours with minimal hands-on time. The CleanPlex® 3-step workflow requires minimal tube-to-tube transfers that can be easily automated on liquid handling platforms.



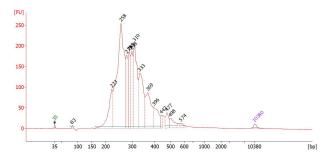
OmniFusion<sup>TM</sup> Target Enrichment and Library Preparation 6.5 hours of total assay time, 85 minutes of hands-on time



### OmniFusion<sup>TM</sup> RNA Lung Cancer Panel | Product Sheet

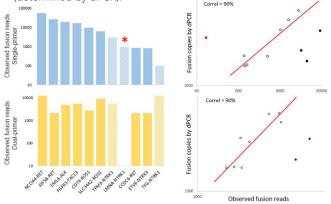
#### CleanPlex® Background Cleaning Chemistry

The OmniFusion™ RNA Lung Cancer Panel is powered by Paragon Genomics' CleanPlex® technology, which uses a proprietary multiplex PCR background cleaning chemistry to effectively remove non-specific PCR products, resulting in best-in-class target enrichment performance and efficient use of sequencing reads.

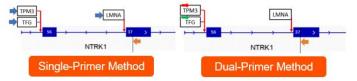


#### Identification of novel and known gene fusions

Seraseq® Fusion RNA mix v4 was used to generate libraries with the OmniFusion<sup>TM</sup> and AccuFusion<sup>TM</sup> RNA Lung Cancer Panels. Not only were the 10 expected fusions in the control material detected with both panels, but one additional novel fusion (LMNA-NTRK1, as indicated by \* below) was also detected with OmniFusion<sup>TM</sup> chemistry (blue bar graph). In addition, the uniform amplification of the CleanPlex® chemistry is highlighted by the high correlation seen between observed reads and fusion copies (determined by dPCR).



Paragon Genomics offers two fusion detection methods: OmniFusion<sup>TM</sup> for the detection of novel fusions, and AccuFusion<sup>TM</sup> for the targeted detection of known fusions. The single-primer method of OmniFusion<sup>TM</sup> is able to identify novel fusions with unknown information (i.e., LMNA-NTRK1 fusion), whereas the dual-primer method of AccuFusion<sup>TM</sup> is used for focused interrogation of known fusion targets (i.e., TPM3/TFG-NTRK1 fusions).



#### OmniFusion<sup>TM</sup> RNA Lung Cancer Panel Performance

|         | Mapping Rate % | On-Target Rate % | rRNA Rate % |
|---------|----------------|------------------|-------------|
| Average | 96.9           | 92.9%            | 0.81        |
| STDV    | 1.0            | 1.7              | 0.27        |

The table above displays the performance of OmniFusion<sup>™</sup> RNA Lung Cancer Panel using 25ng of Seracare® Fusion Reference RNA as input. The generated libraries were sequenced at 0.1 million reads per sample.

## Recommended Sample Multiplexing OmniFusion $^{TM}$ RNA Lung Cancer Panel

| Instrument                               | Samples per Run <sup>A</sup> |
|--|------------------------------|
| iSeq™ i1 System                          | 26                           |
| MiSeq™ System (v2)                       | 98                           |
| MiniSeq™ System (High-output)            | 164                          |
| NextSeq <sup>™</sup> System (Mid-output) | 852                          |
|  |                              |

A. Samples per run at an intended average read depth of 2,500X for 1% MAF detection with 2x 150bp sequencing.

#### Ordering Information

The OmniFusion™ RNA Lung Cancer Panel contains the primers and OmniFusion™ RNA Library Kit. CleanPlex® Indexed PCR Primers and CleanMag® Magnetic Beads are ordered separately to complete the workflow from input RNA to sequencing-ready NGS libraries. For more indexing options and additional product configurations visit www.paragongenomics.com/store/

| Product  | SKU    |
|--|--------|
| OmniFusion <sup>TM</sup> RNA Lung Cancer Panel (8 Rxns)                            | 917100 |
| OmniFusion <sup>TM</sup> RNA Lung Cancer Panel (96 Rxns)                           | 917101 |
| OmniFusion <sup>TM</sup> RNA Lung Cancer Panel (384 Rxns)                          | 917102 |
| CleanPlex® Dual-Indexed PCR Primers for Illumina® Set A (96 indexes, 96 reactions) | 716006 |
| CleanPlex® Dual-Indexed PCR Primers for Illumina® Set B (96 indexes, 96 reactions) | 716018 |
| CleanMag® Magnetic Beads (5 mL)  | 718002 |
| CleanMag® Magnetic Beads (60 mL)   | 718003 |

#### Learn More

To learn more about  $OmniFusion^{TM}$  and  $AccuFusion^{TM}$  technologies, visit

https://www.paragongenomics.com/targeted-sequencing/amplicon-sequencing/rna-fusion-detection/

Paragon Genomics, Inc. | 5020 Brandin Court, FL 2, Fremont, CA 94538, USA | +1.650.822.7545 www.paragongenomics.com | techsupport@paragongenomics.com

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