Genexus System news

Issue: April 30

Note on SARS-CoV-2 testing

We feel it is impossible on this date in April 2020 not to mention the global coronavirus pandemic. Thermo Fisher Scientific is contributing to the fight against the virus all over the world. Researchers around the world are diligently working to gain a better understanding of the coronavirus with the goal of rapidly identifying future treatment options and possible vaccine targets. With a comprehensive portfolio of diagnostic and research products, Thermo Fisher Scientific is your trusted partner for pathogen detection, epidemiological surveillance, vaccine development, and the laboratory equipment needed to advance your research. You can learn more about the end-to-end testing solutions for coronavirus SARS-CoV-2 here.

Specifically for next-generation sequencing (NGS), we have just launched the Ion AmpliSeq™ SARS-CoV-2 Research Panel specifically for next-generation sequencing (NGS)—one assay surveying the entire coronavirus genome for epidemiological investigation. It is currently available for our Ion Torrent™ GeneStudio™ S5 system. To further expedite NGS analysis of SARS-CoV-2 and to help meet growing customer demand, we have begun to optimize the Ion AmpliSeq SARS-COV-2 Research Panel for the Ion Torrent™ Genexus™ System. Optimization and validation of the research panel on the Genexus System is now underway in collaboration with our customers.

“ Quote of the month

The system is basically a fully automated end-to-end workflow that requires very little human input. Therefore, it is very difficult to make any errors, and at the same time, it is very easy to learn how to use it.
–Phillip Jermann, Institute of Medical Genetics and Pathology, University Hospital Basel, Switzerland

Featured publications

What is the real impact?

Experts share their opinion on the value of fully automated NGS results in a single day

The Pathologist Dec 2019

What are the main barriers to NGS adoption in smaller pathology laboratories?

Jose Costa from IPATIMUP says, “The barriers to NGS adoption are the same as for larger laboratories. The difference is the resources to overcome them. In a nutshell, I think it’s the expertise that is needed, from both a molecular biology and a bioinformatics standpoint, including result interpretation. Also, the cost related to both the instruments and the sequencing itself should be taken into account.”

How does the Genexus System help overcome those barriers?

Costa says, “The way the Genexus System overcomes these barriers is essentially because it tackles all these different aspects. The expertise needed to run it is essentially none, because it’s a fully automated system. The machine does everything, including providing a full annotated report for final interpretation. Notably, you no longer need to batch samples. In principle, you can run the system with a single sample. But it is when you have just a handful that it becomes extremely cost-efficient. When I think of costs–not just the reagents, but time needed to process samples–that is a major component.” (continue on page 4).

ThermoFisher SCIENTIFIC

Gene fusion detection by new FusionSync technology

FISH results:

- Formalin-fixed, paraffin-embedded (FFPE) non-small cell lung cancer (NSCLC) sample
- *RET* break-apart probe (32%)

OPA and Genexus results

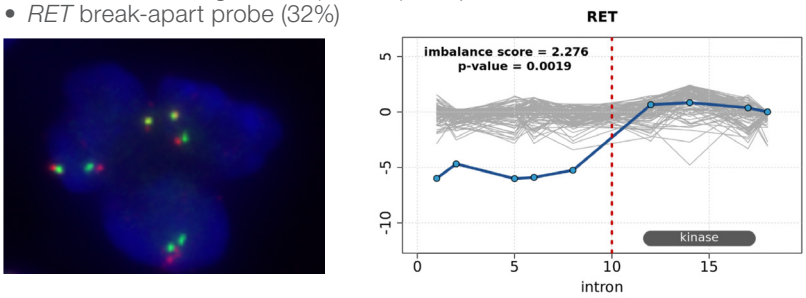


Figure 1. *KIF5B–RET* fusion detected via targeted and imbalance assays.

User classification	Locus	Oncomine variant classification	Genes (exons)	Read counts	Type	Imbalance score	Imbalance *P* value	Predicted breakpoint
--Select--	chr10:32317356 - chr10:43612032	Fusion	***KIF5B*(15)–*RET*(12)**	6887	Fusion	NA	NA	NA
--Select--	chr10:43607628	ExpressionImbalance	**RET**	NA	RNAExonTiles	2.276	0.0019	exon8-exon12

The Oncomine™ Precision Assay was able to confirm the presence of a *RET* fusion in this sample. Additionally, the exon tiling imbalance graph shows a clear difference in expression level across the 3′ and 5′ end of the *RET* gene, along with a statistically significant difference in the expression pattern relative to the *RET* fusion baseline (blue line vs. grey lines). Also, the predicted breakpoint (red dotted line) is consistent with the known location for this fusion. Lastly, the image above shows the original fluorescence *in situ* hybridization (FISH) break-apart probe result, which indicates the presence of a fusion within the *RET* gene.

FISH results:

- Adenocarcinoma FFPE sample
- *NTRK1* break-apart probe (68%)

Oncomine Precision Assay and Genexus System results

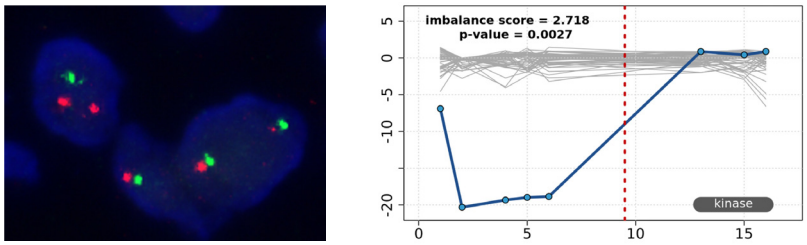


Figure 2. *CLIP1–NTRK1* fusion detected via off-target and imbalance assays.

Gene (exons)	Read counts	Type	Variant ID	Filter	Oncomine driver	Imbalance score	Imbalance *P* value	Predicted breakpoint
NTRK1	NA	RNAExonTiles	N TRK1	PASS	*NTRK1*	2.718	0.0027	Exon 6-exon 13
***CLIP1*(20)–*NTRK1*(11)**	596	Fusion	CLIP1-NTRK1.C20N11. Non-Targeted	PASS	*NTRK1*	NA	NA	NA

The Oncomine Precision Assay was able to confirm the presence of a *NTRK1* fusion in this sample. Additionally, the exon tiling imbalance graph shows a clear difference in expression level across the 3′ and 5′ end of the *NTRK1* gene, along with a statistically significant difference in the expression pattern relative to the *NTRK1* fusion baseline (blue line vs. grey lines). Also, the predicted breakpoint (red dotted line) is consistent with the known location for this fusion. Lastly, the image above shows the original FISH break-apart probe result, which indicates the presence of a fusion within the *NTRK1* gene.

Editor's column

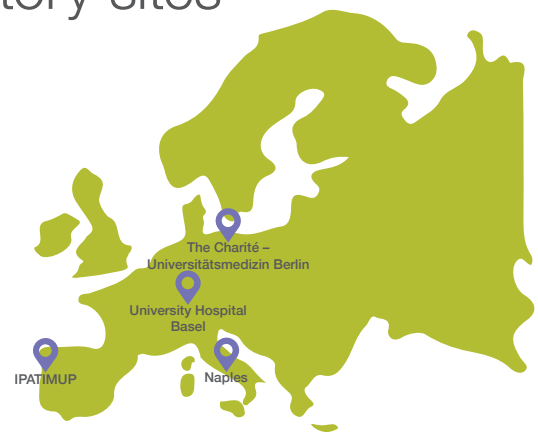
In this issue, we are sharing with you some more data on fusion detection using the new Ion Torrent FusionSync™ detection method, from our customer Basel, Switzerland. The results were generated on real clinical research samples and confirmed with FISH. Overall, these results demonstrate the utility of the Oncomine Precision Assay and FusionSync method in detecting gene fusions that are consistent with other technologies like FISH.

On the third page we show first glimpse of collated data from evaluation of the performance of the Oncomine Precision Assay on the Genexus System in 4 laboratories in Europe. You can see all the data presented by Prof. Michael Hummel from Berlin during our virtual conference - Oncomine World on demand.

Multi-centric evaluation of performance of the Genexus Integrated Sequencer and Oncomine Precision Assay across four external customer laboratory sites

Study overview:

- The following are the sites that contributed to this study: IPATIMUP, University Hospital Basel, The Charité – Universitätsmedizin Berlin, and NT Fondazione Pascale Naples
- Each external customer site selected their own clinical research samples (both FFPE tissue and plasma samples), where a majority of the samples were previously characterized to have a known variant by other assays and/or technologies.
- In total, 74 FFPE samples and 12 plasma samples were analyzed.



Study results:

- Results from this study demonstrated that Ion Torrent™ Genexus™ Integrated Sequencer and Oncomine Precision Assay can detect different variant types (including indels, SNVs, CNVs, and fusions) in both FFPE and plasma samples
- Comparison to previous characterization demonstrated high performance for all variant and sample types
- Implementation and operation of Genexus Integrated Sequencer and Oncomine Precision Assay was successful across four external customer sites with a variety of banked cancer samples

Variant type	Sensitivity	Specificity	Concordance
Indel	95%	100%	95%
SNV	100%	100%	100%
SNV and indel	95%	100%	95%
CNV	100%	100%	100%

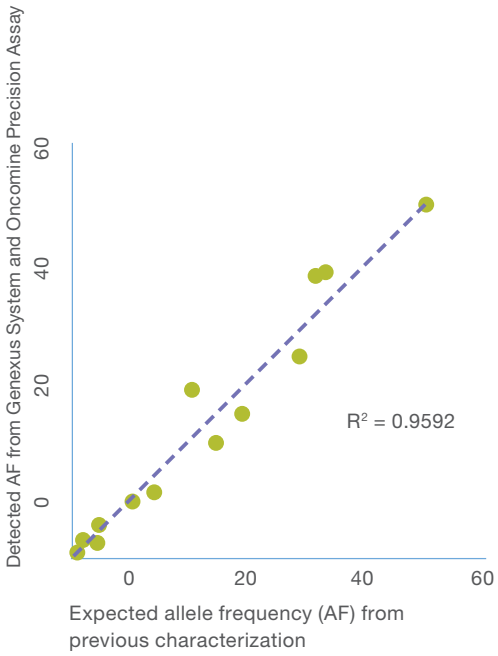


Figure 3. High performance of mutation detection with plasma samples.

The Oncomine Precision Assay has demonstrated high sensitivity, specificity, and overall concordance in detecting mutations (SNV + indel) in plasma samples that were previously characterized with other assays and technologies. Results above included a concordant detection of an *FGFR1* amplification from a CRC sample. Previous characterization included: Ion Torrent™ Oncomine™ Pan-Cancer Cell-Free Assay and Oncomine™ Cell-Free Total Nucleic Acid Research Assay.

Continued from p 1

So labs that don't have any experience with NGS can use the Genexus System?

Phillip Jermann of the Institute of Medical Genetics and Pathology, University Hospital Basel, Switzerland, where NGS was adopted a few years ago, commented, "The Genexus System enables laboratories that want to implement NGS into their routine laboratory workflow and do not yet have the background and the knowledge around NGS to do that easily. That is because the system is basically a fully automated end-to-end workflow that requires very little human input."

And what about experienced laboratories?

Jermann says, "Experienced laboratories like ours can also benefit from the Genexus System because it allows us to automate many steps that are otherwise error-prone when done manually, even by skilled users. So, again, it's the automation that benefits us most. And of course, that goes along with another big advantage of the system–the short turnaround time.

An important aspect of today's NGS based molecular profiling is the percentage of failed tests. Does the Genexus System reduce that percentage?

Jermann says, "Genexus System is based on technology that has evolved over the last 10 years. I was an early adopter, so I have followed its entire development. The method, to the best of my knowledge, has been cited in more than 6,000 scientific papers. Recently, a seminal retrospective study from the University of Heidelberg in Germany that used the Ion Torrent™ technology demonstrated a nearly 97% sequencing success rate across >3,000 NSCLC samples, and a rejection rate of only around 3%. The Genexus System represents a further evolution of that robust methodology, so it is conceivable to expect it will deliver excellent results.

Read the full article here.