Next Generation Sequencing – Applications

Dr Xosé M Fernández
Market Development – Genomics and Bioinformatics, EMEA

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Offering patients hope

Hope: that’s what clinical trials offer to patients around the world who are waiting for promising new treatments and cures for illness and disease. As pharmaceutical and biotech companies work to develop groundbreaking new therapies, they rely on Thermo Fisher Scientific to manage their clinical trials. We’ve supported thousands of trials by packaging and labeling the medicines and ensuring that treatments are properly stored and delivered to patients on time.

We help our customers provide hope to those who are waiting for cures. We are Thermo Fisher Scientific.
Cancer is a Molecular Disease

Change of Paradigm – From Anatomical to Molecular Approach

Breast Cancer
Cervical Cancer
Colorectal Cancer
Liver Cancer
Lung Cancer
Ovarian Cancer
Pancreatic Cancer
Prostate Cancer
Skin Cancer
Stomach Cancer
Thyroid Cancer

ALK
AKT1
BRAF
EGFR
ERBB2
KRAS
NRAS
MAP2K1
PIK3CA
RET
ROS1
Undefined

Source: Nature Medicine, volume 18, number 3, March 2012
### Genomic Research

#### Ever-Expanding List of Genes and Variants

<table>
<thead>
<tr>
<th>Tumor Type</th>
<th>Genes Identified</th>
<th>Driver Mutations</th>
<th>On-Market Drugs</th>
<th>Pipeline Drugs</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lung</td>
<td>80</td>
<td>8-10</td>
<td>2</td>
<td>17</td>
</tr>
<tr>
<td>Breast</td>
<td>47</td>
<td>5-8</td>
<td>1</td>
<td>15</td>
</tr>
<tr>
<td>Colorectal</td>
<td>86</td>
<td>7-10</td>
<td>1</td>
<td>11</td>
</tr>
<tr>
<td>Prostate</td>
<td>93</td>
<td>4-7</td>
<td>0</td>
<td>10</td>
</tr>
</tbody>
</table>

1. Based on q-value analysis using MutSig software from the Broad Institute
2. Based on expert interviews
3. Based on Evaluate Pharmaceuticals database; for pipeline, includes Phase 1 and above only
Ion Torrent™ NGS System coupled with Ion AmpliSeq™ Technology

The answer to the challenges addressing the paradigm change from organ to molecular approach, allowing comprehensive molecular analysis of each sample

One Test, One Result: Multiple biomarkers detected in one test from one sample, in one streamlined workflow

One Technology: detection of different types of biomarkers: hotspots, SNPs, CNVs, indels and gene fusions

Any Sample: From as low as 10 ng of DNA or RNA from FFPE tissue ...
Next Generation Sequencing in Clinical Research

Gene Panels
- Ion AmpliSeq panels
- Oncomine™ Assays

cfDNA Assays
- Observed LOD 0.1%
- Lung
- Colon
- Breast

Genetic Analysis of FFPE samples
- Fast sample-to-answer
- Maximum recovery
- More genetic answers

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Oncology Solid Tumour Biomarker Testing

Today’s Challenges

- Multiple biomarkers for one disease indication
- Multiple type of biomarkers
- Time factor
  - Need to test multiple biomarkers simultaneously
- Limited samples available
- Fixed assay need for validation and standardisation
  - What is the right content?
Expanding Menu of Panels and Assays for Cancer Research

Ion AmpliSeq Panels
- Robust results with limited sample input
- Fast turn-around time
- Pre-designed panels
- Custom-design options

- AmpliSeq Cancer Hotspot Panel v2
  - 50 Genes
  - Complete amplicon coverage of coding exons and exon-intron boundaries

- AmpliSeq Comprehensive Cancer Panel
  - 400 Genes
  - > 70 fusion transcripts

- AmpliSeq Colon and Lung Research Panel v2
  - 22 Genes & Fusions
  - Exons and UTRs

- AmpliSeq BRCA1 and BRCA2 Panel
  - 22 Genes & Fusions

- AmpliSeq RNA Fusion Research Panel
  - > 70 fusion transcripts

- AmpliSeq TP53 Panel
  - Exons and UTRs

- AmpliSeq AML Panel
  - 21 Genes

- AmpliSeq White Glove
  - 22 Genes & Fusions

- AmpliSeq Custom Panels

Oncomine Assays
- Content informed by the Oncomine Knowledgebase
- Based on trusted AmpliSeq Technology
- Single workflow for DNA and RNA
- Developed for use with FFPE Samples
- Protocol based on validation with clinical samples

- Oncomine Comprehensive Assay
  - 143 Genes
  - Hotspots
  - SNVs
  - Indels
  - CNVs
  - Fusions

- Oncomine Focus Assay
  - 52 Genes
  - Hotspots
  - SNVs
  - Indels
  - CNVs
  - Fusions

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Oncomine Assay Workflow™ From Sample to Result and Interpretation

One Sample

FFPE material including fine needle aspirates,
As little as 10ng = Few slides

One test

Flexible and scalable NGS workflow

One Analysis

Streamlined Analysis
Mutations
Copy Number
Variants
Gene Fusions

One Report = Multiple Results

Oncomine Knowledgebase Reporter
Including result Interpretation

For Research Use Only. Not for use in diagnostic procedures.
An easy-to-use interface enables streamlined access to a sample specific variant report in three easy steps.

1. **UPLOAD**
   Investigate biologically relevant variants for cancer research.

2. **FILTER**
   Identify curated content matching source, cancer type and global trial criteria.

3. **REPORT**
   Customize reports for your laboratory.
Generating a Custom Report

Variation Summary

<table>
<thead>
<tr>
<th>Gene</th>
<th>Raw Variant Type</th>
<th>Type</th>
<th>Other Variant Type</th>
<th>Type</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
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</table>

Relevant Therapy Summary

<table>
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<tr>
<th>Gene</th>
<th>Therapy Type</th>
<th>Dose</th>
<th>Route</th>
<th>Other Therapy Type</th>
<th>Dose</th>
<th>Route</th>
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</thead>
<tbody>
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</tbody>
</table>

Current Global Clinical Trials Information

<table>
<thead>
<tr>
<th>Trial</th>
<th>Condition</th>
<th>Disease</th>
<th>Study Type</th>
<th>Stage</th>
<th>Phase</th>
</tr>
</thead>
<tbody>
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A Faster Way to Try Many Drugs on Many Cancers

So-called basket studies, which group cancer patients in a new way, could revolutionize the path from the lab to FDA approval and market success.
Determine whether treating cancers according to their molecular abnormalities (unlike the traditional approach based on treatment guidelines and drugs approved for individual cancer types – e.g. lung, colon...) will show evidence of effectiveness

Demonstrate the use of standardised NGS assay/platform across multiple independent clinical sequencing sites for genomics-informed targeted treatment of cancer patients

Overcome drug development challenges with clinical trial enrolment and patient matching

Avoid undue costs and time-to-enrolment process

Partnership between NCI (National Cancer Institute US) and multiple pharma companies supplying approved and investigational treatments
Our Vision for the Future: Oncomine Universal Dx Product

One workflow to deliver results across many biomarkers enabled by targeted NGS

One Test — all answers for patient stratification to available targeted therapies

Oncomine Focus Assay RUO Early Development

Oncomine Universal Dx Test IUO Registration Trials

Oncomine Universal Dx Test IVD Companion Diagnostics

2015

2016

Solid tumour sample

Oncomine IVD Assay

Sample report

Clinician-patient treatment decision

The content provided herein may relate to products that have not been officially released and is subject to change without notice.

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Oncomine Solid Tumour kits CE IVD

Relevant Content
For guidance of therapeutical decisions
Designed in cooperation with group of leading cancer clinicians: OncoNetwork

Manufactured according to ISO 13485 standards
High level of Quality Control helps provide confidence in reproducibility, accuracy, and consistent quality of results

In compliance with current European IVDD
Helps simplify in-house validation for accreditation purposes

Flexible and Scalable 96 test kit
Consists of 6 packs, 16 tests each and it is possible to run 1-16 samples at once

Multiple results
One Workflow

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Oncomine Solid Tumour Kits

All the answers for therapy management of NSCLC or colon cancer patient from 1 DNA/RNA sample, in one workflow, at the same time

Colon Cancer biomarkers

KRAS
NRAS
BRAF
PIK3CA
PTEN

NSCLC Cancer Biomarkers

EGFR
ALK
ROS1
RET
ERBB2

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Today’s Challenges

- Sample stability
- Standardisation
- Contamination between samples
- Limited material for molecular analysis
- Limit of detection below 1%
- Increase number of target mutations
- False positive rate
NGS cfDNA Assays* – Reinventing How Researchers Target Cancer

**Application – Ion 530 Chip and Ion 540 Chip**
Demonstrated on Proton and Ion PGM Systems

**Critical Hotspots:**
*EGFR:* T790M, L858R, Exon 19 del
*KRAS:* G12X, G13X, Q61X,
*BRAF:* V600E
*ALK:* Exon 21-25,
*PIK3CA:* E545K, H1047R, E542K, and more

**Genes covered:**
*ALK, BRAF, EGFR, ERBB2, KRAS, MAP2K1, MET, NRAS, PIK3CA, ROS1, TP53*

**Tube of blood**
20 ng input – 0.1% LOD
Flexible down to 1 ng

*Data for NGS cfDNA Assays*
NEW! Oncomine Lung cfDNA Assay
LiquidBiopsy™ Platform for CTC and cfDNA isolation
MagMAX™ extraction kits for manual and automated cfDNA isolation
Software tools for rare variant analysis

- Circulating free DNA (cfDNA) isolation
- Circulating tumour cells (CTCs) enrichment
- NGS sequencing assays for both CTC DNA and cfDNA analysis
- Digital PCR
- Unique workflows to get results from both cfDNA and CTCs from one sample, in one analysis
Summary

Next Generation Sequencing is technology of choice for multiple biomarker testing in oncology

Ion Torrent NGS platforms combined with AmpliSeq Technology are proven golden standard allowing robust analysis of FFPE tissue with minimum sample required

Liquid biopsy applications are the new frontiers for cancer research that may potentially impact precision medicine in the future

Thermo Fisher Scientific

is pioneering the way of the NGS use in oncology and constantly developing new applications and solutions

is working with pharma partners towards broad implementation of NGS for selection of patients to clinical trials and for approved treatments
Career Professional Development in Genomic Medicine

- Learn about the main NGS platforms and gain knowledge on the applications of NGS in biomedical research.
- Acquire an understanding of technology implementations revealing disease biology and informing clinical decisions.
- Identify the latest advances in genomics applied to cancer research and the challenges to secure effective interventions based on this new area of scientific investigation.
- Understand current limitations of NGS in clinical practice (with some illustrative examples of analysis workflows) alongside other bioinformatics considerations about these data.
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