Empowering STAT DNA Testing for Molecular Oncology Applications Using A Fully Automated Platform

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Outline

• Human cancer
• How the diagnosis is made
• Precision/personalized medicine in oncology
• The role of molecular testing
• To NGS or not to NGS
• The role of STAT DNA testing
A MESSAGE OF HOPE

CANCER is a curable disease. CANCER is neither contagious nor hereditary. Yearly 90,000 people (1 in 10 over 40 years old) die of this disease in this country. Many of these victims could have been cured had they gone to a reputable doctor immediately. “Immediately” means as soon as symptoms are noticed.

Shown for the American Society for the Control of Cancer—A Benevolent Organization.
370 Seventh Avenue, New York City.
### Estimated New Cases

<table>
<thead>
<tr>
<th>Site</th>
<th>Males</th>
<th>Females</th>
<th>Percentage</th>
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<td>Prostate</td>
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<td>Urinary bladder</td>
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<td>Melanoma of the skin</td>
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<td>Non-Hodgkin lymphomas</td>
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<td><strong>All Sites</strong></td>
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<table>
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<tr>
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<td>Non-Hodgkin lymphoma</td>
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<tr>
<td>Pancreas</td>
<td>26,240</td>
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<tr>
<td>Leukemia</td>
<td>25,270</td>
<td>3%</td>
<td></td>
</tr>
<tr>
<td>Kidney &amp; renal pelvis</td>
<td>22,660</td>
<td>3%</td>
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</tr>
<tr>
<td><strong>All Sites</strong></td>
<td>878,989</td>
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### Estimated Deaths

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<tr>
<td>Lung &amp; bronchus</td>
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<tr>
<td>Prostate</td>
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<td>Pancreas</td>
<td>23,020</td>
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<tr>
<td>Liver &amp; intrahepatic bile duct</td>
<td>20,540</td>
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<tr>
<td>Leukemia</td>
<td>14,270</td>
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<tr>
<td>Esophagus</td>
<td>12,850</td>
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<tr>
<td>Urinary bladder</td>
<td>12,520</td>
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<tr>
<td>Non-Hodgkin lymphomas</td>
<td>11,510</td>
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<tr>
<td>Kidney &amp; renal pelvis</td>
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<tr>
<td><strong>All Sites</strong></td>
<td>323,630</td>
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<table>
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<th>Site</th>
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<td>Lung &amp; bronchus</td>
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<td>Breast</td>
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<td>Colon &amp; rectum</td>
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<td>Pancreas</td>
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<td>Ovary</td>
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<td>Uterine corpus</td>
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<td></td>
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<tr>
<td>Leukemia</td>
<td>10,100</td>
<td>4%</td>
<td></td>
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<tr>
<td>Liver &amp; intrahepatic bile duct</td>
<td>9,660</td>
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<tr>
<td>Non-Hodgkin lymphoma</td>
<td>8,400</td>
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<tr>
<td>Brain &amp; other nervous system</td>
<td>7,340</td>
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<tr>
<td><strong>All Sites</strong></td>
<td>286,010</td>
<td>100%</td>
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</tr>
</tbody>
</table>
1. Unregulated (clonal) cell growth
2. Impaired cellular differentiation
3. Invasiveness
4. Metastatic potential
Human Cancer as a Genetic Disease

Cancer results from the disruption of important genes and gene products.
Human Cancer as a Genetic Disease

• ~30,000 genes in human genome
• Only a small fraction of these genes have the potential to cause cancer when mutated
  • Oncogenes
  • Tumor Suppressor Genes
Human Cancer as a Genetic Disease
Human Cancer – The Diagnosis

H&E for Morphology

Further Diagnostic Work-up
- Imaging (CT, MRI, PET)
- Immuno (IHC), molecular,

1.4M New Cancers (US)

Therapeutic management

[Imagery of cancer cells and diagnostic tools]
Human Cancer – The Diagnosis
Human Cancer – The Diagnosis

Nothing Else Looks Like This!
Promises of the Human Genome

- Diagnostic
- Prognostic
- Predictive
- Therapeutic
Human Cancer - Precision Medicine

• >2 million ADRs occur annually in US
• ~100,000 deaths (4th leading cause of death)
• >$76 billion - cost of drug-related morbidity & mortality
• 4% of new drugs are withdrawn due to ADRs
  • 1995-2005: 34 drugs withdrawn mainly due to hepatotoxic or cardiotoxic effects
• Therapeutics effective in 25-60% of patients
• Genetics accounts for ~24% of drug disposition and effects.
  • Due to polymorphisms in drug metabolizing enzymes, transporters, and targets (receptors)
Human Cancer - Precision Medicine

• PGx\textsubscript{m}: pharmacokinetic
  • What the body does to the drug
    • Absorption
    • Distribution
    • Metabolism
    • Excretion

• PGx\textsubscript{t}: targeted therapy
  • Presence/absence of therapeutic target
  • Response or lack of response
  • Resistance
  • Local or distant recurrence
Human Cancer - Precision Medicine

• PGx\textsubscript{m}: pharmacokinetic
  • Polymorphisms
  • Not typically disease causing mutations
  • Ex. Irinotecan and UGT1A1

• PGx\textsubscript{t}: targeted therapy
  • Mostly mutations in disease causing genes
  • Includes driver and passenger mutations
  • Germline vs somatic variants
Human Cancer - Precision Medicine PGxₘ

Genetic polymorphisms

Spectrum of Drug Metabolism

- Poor (PM)
- Intermediate (IM)
- Extensive (EM)
- Ultra-Rapid (UM)
Human Cancer – Targeted Therapy (PGX$_t$)

- EGFR inhibitors
- Cyclin-dependent kinase inhibitors
- Immune activating anti-CTLA4 mAb
- Telomerase Inhibitors
- Selective anti-inflammatory drugs
- Inhibitors of VEGF signaling
- Inhibitors of HGF/c-Met

Aerobic glycolysis inhibitors
- Resisting cell death
- Deregulating cellular energetics
- Sustaining proliferative signaling
- Evading growth suppressors
- Avoiding immune destruction
- Enabling replicative immortality
- Tumor-promoting inflammation
- Inducing angiogenesis
- Activating invasion & metastasis
- Genome instability & mutation
- Proapoptotic BH3 mimetics

Human Cancer – Targeted Therapy (PGXₜ)

- BCR-ABL1
  - Imatinib (Gleevec) for CML
- HER2 amplification
  - Trastuzumab (Herceptin) for breast cancer
- KRAS point mutation
  - Cetuximab and Panitumimab for colon cancer
- EGFR point mutation and/or amplification
  - Iressa, Tarceva for lung cancer
Wild-Type KRAS Is Required for Panitumumab Efficacy in Patients With Metastatic Colorectal Cancer

Rafael G. Amado, Michael Wolf, Marc Peeters, Eric Van Cutsem, Salvatore Siena, Daniel J. Freeman, Todd Juan, Robert Sikorski, Sid Suggs, Robert Radinsky, Scott D. Patterson, and David D. Chang
Targeting the EGFR Pathway in NSCLC

- EGFR mutations and sensitivity to TKIs
  - Exon 19 deletion
  - Exon 21 (L858R)
  - Exon 18 (G719X)

- Exon 20 insertion may predict resistance to TKIs
- EGFR and KRAS mutations are mutually exclusive in NSCLC
- KRAS mutation associated with primary resistance to TKIs
The Role of Molecular Dx in Oncology
(Somatic Mutation Detection)

EGFR Exon 19 Deletion Analysis

9bp deletion

12bp deletion

15bp deletion

18bp deletion

EGFR Exon 21 SNP (L858R) Analysis

KRAS Analysis x7
The Role of Molecular Dx in Oncology
(Sanger Sequencing - Somatic Mutation Detection)

1 ctccgggtg tcccgctcg gcaagcgctg cccaggtctg ggggtgtggg cagccagcgg
61 gacagggaa ggaagatgt tcccgctcg cccaggtctg tgggtctggg tgtcttggg
121 caccagctgg gtaggctggg ggaagctcct gcagagcttg gacaggaaga ggcagttgta
181 cgtggctct caggccatc gttggagcta ccgacgtcct cccacaaact caaggtcctg
241 tctctctgta acctctctta agaaaattgt ctacagagac tagaaccata attttaaga
301 agagaaaaac caaatctgct ttcagagact cgtgtggcct acatattatg ctgaagttcgtg
361 agacacatcata aagttctact ttaaaaataa ggcaagataag cccttggagca tccatctctca
421 aggaattagg tacagtaaat tcatcagag tggctcttac ctggacacactc catccctctc
481 agagacagatg gacacgcttg tggctcag ggagctttcat tctacatatg ggagctgctg
541 tgagacagct gacaccagcc atgtgcctc tccatgcctc acacactct atacttccca
601 tgaatatctg atcgagatt tcatcactgc cccttggagct cttggttaaaa
661 agggacccatg actgaggggtg ggacacagaga gacgtttgac aagcaatctg tgctactattt
721 tgctgtgttt gatgaaagca aggctggcct cacgctcact tctcttctgct cttggttaaaa
781 ttgatattg aatgggacac tggccagata aacagttgtg gccttcagac acatcactg
841 gcacagctgt ggaatgacgt ctggccagct cattctctc tcattctcga acggtctggtt
901 cttggagcc aaccatatcattgcacttcg catcaccctt gtcagtccta cattcactac
961 cgcaaatatg actgtgctcg cagaggaata tgtcgatcata cttctctca ccccaaaaaca
The Role of Molecular Dx in Oncology
(Somatic Mutation Detection)

• Single gene assays
• Single variants
• Labor intensive
• Costly
• Algorithms for testing
• Increasing demand
• Increasing numbers of genes and variants
The Role of Molecular Dx in Oncology
(Next Generation or Massively Parallel Sequencing - Somatic Mutation Detection)

• Low quantities of DNA
• Multiple genes (10-380 genes) simultaneously
• Each DNA fragment sequenced 100’s-1,000’s x
• Multiple patients’ samples (8-10) simultaneously
The Role of Molecular Dx in Oncology
(NGS - Somatic Mutation Detection)
### Somatic Mutation Analysis (NGS)

Ion Torrent Cancer Hotspot v2 gene panel (CHPv2) (50)

<table>
<thead>
<tr>
<th>Gene 1</th>
<th>Gene 2</th>
<th>Gene 3</th>
<th>Gene 4</th>
<th>Gene 5</th>
</tr>
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<tbody>
<tr>
<td>ABL1</td>
<td>EGFR</td>
<td>GNAS</td>
<td>KRAS</td>
<td>PTPN11</td>
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<tr>
<td>AKT1</td>
<td>ERBB2</td>
<td>GNAQ</td>
<td>MET</td>
<td>RB1</td>
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<tr>
<td>ALK</td>
<td>ERBB4</td>
<td>HNF1A</td>
<td>MLH1</td>
<td>RET</td>
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<tr>
<td>APC</td>
<td>EZH2</td>
<td>HRAS</td>
<td>MPL</td>
<td>SMAD4</td>
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<tr>
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<td>FBXW7</td>
<td>IDH1</td>
<td>NOTCH1</td>
<td>SMARCB1</td>
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<tr>
<td>BRAF</td>
<td>FGFR1</td>
<td>IDH2</td>
<td>NPM1</td>
<td>SMO</td>
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<tr>
<td>CDH1</td>
<td>FGFR2</td>
<td>JAK2</td>
<td>NRAS</td>
<td>SRC</td>
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<tr>
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<td>PDGFRA</td>
<td>STK11</td>
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<tr>
<td>CSF1R</td>
<td>FLT3</td>
<td>KDR</td>
<td>PIK3CA</td>
<td>TP53</td>
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<tr>
<td>CTNNB1</td>
<td>GNA11</td>
<td>KIT</td>
<td>PTEN</td>
<td>VHL</td>
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</table>

(207 amplicons, >20kb, 10ng DNA input)

SPECIAL ARTICLE
Guidelines for Validation of Next-Generation Sequencing Based Oncology Panels: A Joint Consensus Recommendation of the Association for Molecular Pathology and College of American Pathologists (J Molec Diagn 2017)

Lawrence J. Jennings, Maria E. Arcila, Christopher Corless, Suzanne Kamel-Reid, Ira M. Lubin, John Pfeifer, Robyn L. Temple-Smolkin, Karl V. Voelkerding, and Marina N. Nikiforova
The Role of Molecular Dx in Oncology
(NGS - Somatic Mutation Detection)

Molecular testing ordered by surgical pathologist
2 H&E and 10 USS

MG Pathologist review of H&E for adequacy and % tumor

1 H&E and 2 USS to FISH lab to hold for additional testing if needed

DNA/RNA extracted from USS in molecular laboratory
To NGS or Not To NGS

**Sample Preparation**
- DNA Extraction: minimum tumor cellularity: 10% - 8 unstained slides
- DNA Quantification: PicoGreen Method

**Library Preparation**
- PCR AmpliSeq HotSpot Cancer Panel: 201 amplicons, 50 genes, require 10ng DNA
- FuPa Treatment
- Barcode Adaptor Ligation
- Library Quantification and Pooling (qPCR)

**Emulsification and Enrichment**
- Emulsification PCR: clonal amplification of DNA on Ion Spheres (ISP’s)
- ISP’s quantification
- Enrichment of ISP’s with DNA

**Sequencing and Data Analysis**
- 318 IonChip: Majority of amplicon coverage >500X
- Variant Calling: Ion Torrent Variant Caller Plugin, Reference genome: hg19
- Reporting: Golden Helix SVS Software, Variant Call Summary, Variant Prediction
- Data Annotation, Review and Sign-out

**Times**
- Total time: ~9h
- Hands on time: ~3h
- Total time: ~8h
- Hands on time: ~4h
- Total time: ~7h
- Hands on time: ~1h
- Total time: ~14h
- Hands on time: ~5h
To NGS or Not To NGS

Complexity of Somatic Mutation Analysis

• Clinically actionable (sensitizing or resistance) and FDA approved application

• Clinically actionable but off label (drug not approved for tumor type, maybe for compassionate use)

• Clinically actionable to select clinical trial

• Not actionable but therapeutics in the pipeline
To NGS or Not To NGS

Complexity of Somatic Mutation Analysis

• How many genes and which ones do we really need to test
• Which mutations are most important
• Which combinations of mutations may be important
• Are we treating the 5-20% tumor cells with mutation or the 80-95% without
• What about the 10% of cases that are wild type
• Regulatory and reimbursement issues
ABSTRACT (1998) - DNA STAT

Gregory J. Tsongalis

Introduction. Rapid advances in molecular biology techniques over the past few years have resulted in a transition of these technologies from the research laboratory to the clinical laboratory and in the near future to the bedside. Following in the footsteps of other more established clinical diagnostic technologies, nucleic acid testing is becoming automated and very routine for the evaluation of hematologic, infectious, and genetic diseases. One disadvantage of these new technologies has been the inability for rapid turn around times, a clinical assay attribute crucial for the critically ill patient. While a STAT designation is unbecoming of nucleic acid based tests, new methods for performing DNA/RNA extraction, amplification and detection have reduced the turn around times for these assays dramatically. The aim of this study is to demonstrate some of the time savings in performing nucleic acid tests based on currently available technologies with respect to assays suitable for the critical care patient.

Methods. Random whole blood specimens which were submitted for CBCs were received from Hematology. DNA extraction was performed using the Puregene Kit (Gentra Systems, Minneapolis, MN) according to the recommendations of the manufacturer. Multiple PCR assays were evaluated for different target sequences, including human genomic targets and microbial targets in a time study to optimize amplification efficiency and turn around times. Detection methods included agarose and polyacrylamide gel electrophoresis, liquid hybridization assays, and fragment size analysis using an automated DNA sequencing system (OpenGene, Visible Genetics, Toronto, Canada).

Results. Using rapid column extraction protocols, DNA suitable for PCR amplification can be isolated from whole blood specimens in less than 30 minutes. While PCR amplification times are most often target dependent, newer thermal cyclers can speed this process to less than two hours. Detection by gel electrophoresis, liquid hybridization and/or automated DNA sequencer analysis can also be accomplished within two to three hours. Thus, a completed molecular diagnostic assay for the qualitative detection of a target sequence can be accomplished with an approximately five hour turn around time.

Conclusions. In this study, we demonstrate the feasibility of a STAT nucleic acid based test. Using modified protocols and newer technologies, we are able to detect the presence of a target sequence within five hours. While five hours may not seem appropriate for a STAT designation with respect to more traditional automated clinical diagnostic assays, this is extremely rapid for a molecular based assay. However, with respect to the critical care patient, our ability to detect the presence of a microbial pathogen within a few hours versus a few days may prove crucial to decreasing morbidity and mortality of these patients. In addition, continued advances in these technologies such as DNA chip based assays and highly automated instrumentation will continue to drive turn around times downward while maintaining extraordinarily high sensitivities and specificities.
Human Cancer – The Diagnosis
STAT DNA Testing for Oncology?

- Clinical utility
- Complex specimen (FFPE tissue)
- Assay performance
- TAT
- Data analysis
STAT DNA Testing for Oncology

Simplifying FFPE Somatic Mutation Testing
Testing Steps Within the Cartridge

- **Sample liquefaction**
  - Liquifaction buffer/lysis buffer
  - High intensity focused ultrasound
  - Heating
  - Enzymatic

- **Cell lysis**

- **DNA/RNA extraction**
  - DNA fixed on silica membrane
  - Washing steps
  - Elution step

- **Amplification/detection**
  - 5 independent chambers
  - 6 fluorophores per chamber

- **Data analysis**
  - Automated
Amplicons are detected in real time by an allele-specific PlexZyme®.
In a multiplex reaction, the universal probes are labelled with different fluorophores so that fluorescence signal corresponding to detection of each target sequence can be monitored simultaneously in real time. The highly multiplex nature of *PlexZyme* enzymes can maximize the outputs of qPCR instruments.
Idylla Assays Evaluated

• Idylla KRAS Mutation Assay
  • 21 mutations in KRAS exon 2, 3, and 4

• Idylla NRAS-BRAF-EGFR S492R Mutation Assay (NRAS$_3$)
  • 25 mutations in NRAS exon 2, 3, 4, BRAF exon 15, and EGFR exon 12
Validation of Cartridge Based Assays

• Limit of Detection – obtain Horizon FFPE controls that contain cell lines with varying allele frequencies for mutations (ideally 10% or less) and run 5-10 cartridges on the same sample.

• Precision – use the data from the LOD studies in #1 to show that the results are reproducible from run to run and operator to operator.

• Accuracy – using purchased control FFPE material or previously tested patient samples, run 5-10 samples and assess concordance with previous method.

• ****include different types of variants that assay tests for*****
Samples Analyzed

• Colorectal cancer FFPE tissue samples with mutation in *KRAS* (n=17), *NRAS* (n=5), or *BRAF* (n=12) were analyzed (total = 34).

• 10 colorectal cancer tissue samples with no mutation.

• 9 horizon control samples in triplicate (27).

• A single 10 µm FFPE tissue section was used (total of 4 sections and 2 H&E slides obtained from each sample).

• Results were compared against those previously obtained by NGS using the AmpliSeq 50-gene Cancer Hotspot Panel.
## KRAS Results

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<th>Tumor Content (%)</th>
<th>NGS</th>
<th>Idylla</th>
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<tbody>
<tr>
<td>1</td>
<td>10</td>
<td>c.34G&gt;T, p.G12C</td>
<td>c.34G&gt;T, p.G12C</td>
</tr>
<tr>
<td>2</td>
<td>25</td>
<td>c.34G&gt;T, p.G12C</td>
<td>c.34G&gt;T, p.G12C</td>
</tr>
<tr>
<td>3</td>
<td>75</td>
<td>c.35G&gt;A, p.G12D</td>
<td>c.35G&gt;A, p.G12D</td>
</tr>
<tr>
<td>6</td>
<td>30</td>
<td>c.35G&gt;T, p.G12V</td>
<td>c.35G&gt;T, p.G12V</td>
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<tr>
<td>7</td>
<td>60</td>
<td>c.35G&gt;T, p.G12V</td>
<td>c.35G&gt;T, p.G12V</td>
</tr>
<tr>
<td>8</td>
<td>80</td>
<td>c.35G&gt;T, p.G12V</td>
<td>c.35G&gt;T, p.G12V</td>
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</tbody>
</table>
NRAS Results

<table>
<thead>
<tr>
<th>Sample</th>
<th>Tumor Content (%)</th>
<th>NGS</th>
<th>Idylla</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>85</td>
<td>c.35G&gt;T, p.G12V</td>
<td>c.35G&gt;T, c.35G&gt;T, p.G12A/V</td>
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<tr>
<td>3</td>
<td>50</td>
<td>c.183A&gt;C, p.Q61H</td>
<td>c.183A&gt;C; c.183A&gt;T, p.Q61H</td>
</tr>
<tr>
<td>4</td>
<td>40</td>
<td>c.183A&gt;T, p.Q61H</td>
<td>c.183A&gt;C; c.183A&gt;T, p.Q61H</td>
</tr>
<tr>
<td>5</td>
<td>80</td>
<td>c.183A&gt;T, p.Q61H</td>
<td>c.183A&gt;C; c.183A&gt;T, p.Q61H</td>
</tr>
<tr>
<td>Sample</td>
<td>Tumor Content (%)</td>
<td>NGS</td>
<td>Idylla</td>
</tr>
<tr>
<td>--------</td>
<td>-------------------</td>
<td>----------------------------</td>
<td>----------------------------------------------------------</td>
</tr>
<tr>
<td>1</td>
<td>60</td>
<td>c.1799T&gt;A, p.V600E</td>
<td>c.1799T&gt;A; c.1799_1800delinsAA/c.1799_1800delinsAC, p.V600E/D</td>
</tr>
<tr>
<td>2</td>
<td>50</td>
<td>c.1799T&gt;C, p.V600E</td>
<td>c.1799T&gt;A; c.1799_1800delinsAA/c.1799_1800delinsAC, p.V600E/D</td>
</tr>
<tr>
<td>4</td>
<td>60</td>
<td>c.1799T&gt;A, p.V600E</td>
<td>c.1799T&gt;A; c.1799_1800delinsAA/c.1799_1800delinsAC, p.V600E/D</td>
</tr>
<tr>
<td>5</td>
<td>50</td>
<td>c.1799T&gt;A, p.V600E</td>
<td>c.1799T&gt;A; c.1799_1800delinsAA/c.1799_1800delinsAC, p.V600E/D</td>
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<tr>
<td>7</td>
<td>60</td>
<td>c.1799T&gt;A, p.V600E</td>
<td>c.1799T&gt;A; c.1799_1800delinsAA/c.1799_1800delinsAC, p.V600E/D</td>
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</tbody>
</table>
# Horizon Control Results

<table>
<thead>
<tr>
<th>Mutation</th>
<th>Tumor Content (%)</th>
<th>Repeats</th>
<th>Idylla</th>
</tr>
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<tbody>
<tr>
<td>KRAS G12V</td>
<td>50</td>
<td>3</td>
<td>c.35G&gt;T, p.G12V</td>
</tr>
<tr>
<td>KRAS G13D</td>
<td>50</td>
<td>3</td>
<td>c.38G&gt;A, p.G13D</td>
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<tr>
<td>KRAS Q61H</td>
<td>50</td>
<td>3</td>
<td>c.183A&gt;C / c.183A&gt;T, p.Q61H</td>
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<tr>
<td>KRAS A146 T</td>
<td>50</td>
<td>3</td>
<td>c.436G&gt;C / c.436G&gt;A / c.437 C&gt;T, p.A146P/T/V</td>
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<tr>
<td>NRAS Q61H</td>
<td>50</td>
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<td>c.183A&gt;C, p.Q61H</td>
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<tr>
<td>NRAS Q61L</td>
<td>50</td>
<td>3</td>
<td>c.182A&gt;T, p.Q61L</td>
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<tr>
<td>NRAS Q61R</td>
<td>50</td>
<td>3</td>
<td>c.182A&gt;G, p.Q61R</td>
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<tr>
<td>BRAF V600E</td>
<td>50</td>
<td>3</td>
<td>c.1798T&gt;A; c.1799_1800delinsAA/c.1799_1800delinsAC, p.V600E/D</td>
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<tr>
<td>BRAF V600R</td>
<td>50</td>
<td>3</td>
<td>c.1798_1799 delinsAA/c.1798_1799delinsAG, p.V600K/R</td>
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</table>
### A

<table>
<thead>
<tr>
<th>KRAS GENOTYPE</th>
<th>MUTATION DETECTED IN KRAS CODON 13</th>
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<tbody>
<tr>
<td>Mutation</td>
<td>G13D</td>
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<tr>
<td>Protein</td>
<td>p.Gly13Asp</td>
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<tr>
<td>Nucleotide Change</td>
<td>c.38G&gt;A</td>
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### B

<table>
<thead>
<tr>
<th>NRAS GENOTYPE</th>
<th>MUTATION DETECTED IN NRAS CODON 61</th>
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<tbody>
<tr>
<td>Mutation</td>
<td>Q61H</td>
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<tr>
<td>Protein</td>
<td>p.Gln61His</td>
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<tr>
<td>Nucleotide Change</td>
<td>c.183A&gt;C; c.183A&gt;T</td>
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<tr>
<td>BRAF GENOTYPE</td>
<td>NO MUTATION DETECTED IN BRAF CODON 600</td>
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<tr>
<td>EGFR GENOTYPE</td>
<td>NO MUTATION DETECTED IN EGFR CODON 492</td>
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### C

<table>
<thead>
<tr>
<th>NRAS GENOTYPE</th>
<th>NO MUTATION DETECTED IN NRAS CODON 12,13,59,61,117,146</th>
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<tbody>
<tr>
<td>BRAF GENOTYPE</td>
<td>MUTATION DETECTED IN BRAF CODON 600</td>
</tr>
<tr>
<td>Mutation</td>
<td>V600E/D</td>
</tr>
<tr>
<td>Protein</td>
<td>p.Val600Glu / p.Val600Asp</td>
</tr>
<tr>
<td>Nucleotide Change</td>
<td>c.1799T&gt;A; c.1799_1800delinsAA / c.1799_1800delinsAC</td>
</tr>
<tr>
<td>EGFR GENOTYPE</td>
<td>NO MUTATION DETECTED IN EGFR CODON 492</td>
</tr>
</tbody>
</table>
KRAS Detection in Colonic Tumors by DNA Extraction From FTA Paper: The Molecular Touch-Prep

Melissa L. Petras, Joel A. Lefferts, Brian P. Ward, Arief A. Suriawinata, Gregory J. Tsongalis

DOI: 10.1097/PDM.0b013e318211d554

PMID: 22089345
Issn Print: 1052-9551
Publication Date: 2011/12/01
Potential of STAT Somatic Mutation Testing at Resection

M. Rabie Al-Turkmani, Shannon N. Schutz, Gregory J. Tsongalis

Clinical Chemistry 64:5
STAT DNA Testing for Oncology

- Robust performance
- Rapid TAT
- Ease of use
- Molecular touch prep
- Targeted mutations but ALL are actionable
- Potential for liquid bx analysis
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- Leanne Cook
- Sophie Deharvengt
- Betty Dokus
- Torrey Gallagher
- **Kelley Godwin**
- Donald Green
- Cameron Griffin
- Arnold Hawk
- Brianna Houde
- Edward Hughes
- Michael Johnston
- Kathryn Kearns
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- **M. Rabie Al-Turkmani**

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- Sarah Benware – UNH
- Jamie Dinulos – Dartmouth
- Mackenzie Keegan – Northeastern
- Michael Suriawinata - Dartmouth

### CGAT-Admin
- Heather Steinmetz
- Amber Erskine
- Greg Tsongalis
- Wendy Wells

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Please note:

The Biocartis Idylla™ instrument and console are approved for IVD use while the oncology cartridges are for research use only in the United States.

NGS technologies are for research use only.