Real-World Data: Importance of Monitoring KRAS Mutations in Blood

Yishan Chuang, Natalie Todd, Theo Petropolis, Hannah Allen and Joanna Roder.

Biodesix, Inc., Boulder, CO

BACKGROUND

KRAS mutations are known to be associated with a poorer prognosis for patients with non-small cell lung cancer (NSCLC) and until recently, there was no treatment targeted at KRAS mutations. With the promising results of KRAS targeted therapies in clinical studies demonstrating efficacy in the KRAS mutation positive population, it has become apparent that there is a need for swift, blood-based KRAS mutation testing. GeneStrat® mutation testing uses a blood draw to provide molecular results for KRAS G12C/V/D mutations within 72 hours, expediting time to treatment. This study reports on the analytical, clinical, and real-world performance of testing for KRAS mutations using the GeneStrat test for patients with all stages of NSCLC enrolled in the INSIGHT observational study (NCT03289780).

METHODS

The INSIGHT study includes 33 US sites having enrolled 2700 patients to date in a registry-style design allowing NSCLC patients with any stages of disease or lines of therapy to enroll. Patient characteristics, therapeutic decisions, staging, disease monitoring metrics, and available biomarker data are being collected and patients are being followed for 18 months. Patients are receiving blood-based GeneStrat® genomic and VeriStrat® proteomic testing prior to therapy initiation. Circulating tumor DNA (ctDNA) analysis was performed using Droplet Digital™ PCR (ddPCR) in the Biodesix® CAP/CLIA/NYS CLEP/ISO13485-approved lab in Boulder, CO. An interim analysis of the INSIGHT patients with at least one year of follow up was performed. A total of 980 patients were included in the INSIGHT interim analysis.

Average turnaround time was 34.7 hours for the INSIGHT interim analysis

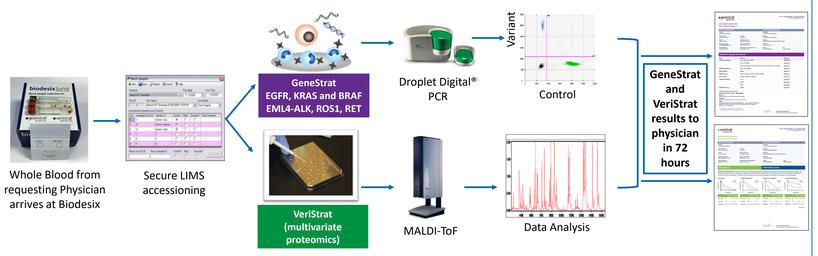


Fig.1 Overview of the current GeneStrat and VeriStrat CAP/CLIA/NYS CLEP Test Workflows. Patient sample testing is initiated when whole blood drawn into blood collection tubes (GeneStrat) or dried serum collection devices (VeriStrat) arrive at the Biodesix Laboratory. According to the test type requested, samples are processed through parallel workflows to isolate circulating DNA and RNA, or to recover serum proteins. For ddPCR analysis, samples are processed using the Bio-Rad QX200 ddPCR system and droplet counts are evaluated using QuantaSoft. For MALDI-ToF analysis, samples are processed on an AutoFlex MS and spectra are analyzed using the VeriStrat classification algorithm.

RESULTS

ANALYTICAL & CLINICAL VALIDATION[1]

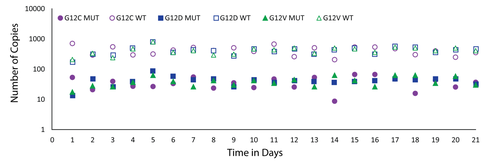


Fig 2. Robustness Testing. Analytical positive control was spiked into normal human plasma, extracted, and tested by Droplet Digital PCR (Bio-Rad, Pleasanton, CA) over 21 consecutive business days. Both mutant (MUT) and wild-type (WT) copies are reported for KRAS variants G12C, G12D, and G12V.

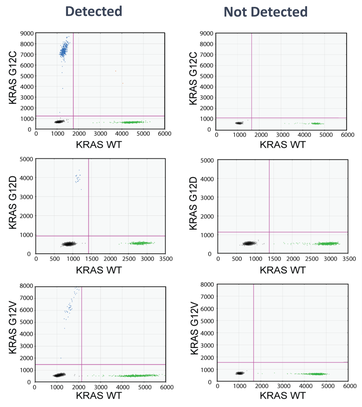


Table 1. Clinical Validation. Summary of sensitivity, specificity, and concordance of KRAS variants G12C, G12D, and G12V, as well as the test performance of the three variants combined from the GeneStrat validation studies. Reference methodology was performed with Therascreen (EGFR and KRAS) from Qiagen.

Variant	Sensitivity	Specificity	Concordance
All KRAS	87.9%	100%	96.0%
G12C	87.5% (7/8)	100% (34/34)	97.6% (41/42)
G12D	78.6% (11/14)	100% (13/13)	88.9% (24/27)
G12V	100% (11/11)	100% (19/19)	100% (30/30)

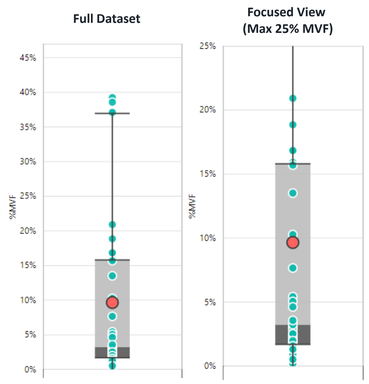
Fig. 3. Clinical Validation. Example of droplet distributions for positive donor samples for each of the KRAS variants evaluated in the GeneStrat validation studies.

INSIGHT STUDY RESULTS

Table 2. Summary of KRAS prevalence in the INSIGHT interim analysis patient population.

	MUT	WT	%
KRAS (G12C/V/D)	**87**	**760**	**10.3%**
Stage IA	1	40	2.4%
Stage IB	0	30	0.0%
Stage IIA	1	31	3.1%
Stage IIB	1	23	4.2%
Stage IIIA	10	101	9.0%
Stage IIIB	5	68	6.8%
Stage IV	69	467	12.9%
KRAS G12C	**41**	**809**	**4.8%**

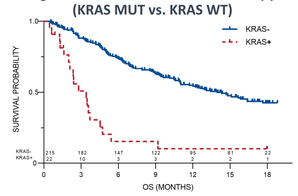
Fig 4. %MVF of KRAS G12C positive results %MVF (percentage of minor variant frequency) was measured in patients from the INSIGHT interim analysis population. Full dataset represent the full range of the %MVF collected from the INSIGHT interim KRAS G12C MUT population (n-43). The focused view excluded patients that have %MVF above 25%.



Median % MVF	Max of % MVF	Min of % MVF
3.40%	39.24%	0.19%

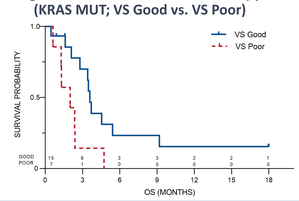
RESULTS (Continued)

Fig 5A. Platinum-Based Chemotherapy (KRAS MUT vs. KRAS WT)



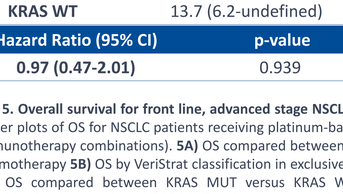
KRAS Status	Median OS (95% CI) in months
KRAS MUT	3.4 (2.0-4.5)
KRAS WT	14.0 (11.0-17.6)
Hazard Ratio (95% CI)	**p-value**
4.27 (2.56-7.10)	<0.001

Fig 5B. Platinum-Based Chemotherapy (KRAS MUT; VS Good vs. VS Poor)



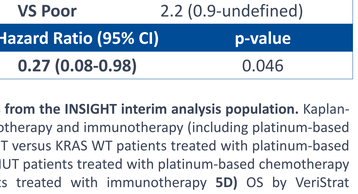
VS Result	Median OS (95% CI) in months
VS Good	3.6 (2.1-5.4)
VS Poor	2.0 (0.6-2.4)
Hazard Ratio (95% CI)	**p-value**
0.32 (0.11-0.88)	0.027

Fig 5C. Immunotherapy (KRAS MUT vs. KRAS WT)



KRAS Status	Median OS (95% CI) in months
KRAS MUT	Not reached (1.9 – undefined)
KRAS WT	13.7 (6.2-undefined)
Hazard Ratio (95% CI)	**p-value**
0.97 (0.47-2.01)	0.939

Fig 5D. Immunotherapy (KRAS MUT; VS Good vs. VS Poor)



VS Result	Median OS (95% CI) in months
VS Good	Not reached (2.2-undefined)
VS Poor	2.2 (0.9-undefined)
Hazard Ratio (95% CI)	**p-value**
0.27 (0.08-0.98)	0.046

Fig. 5. Overall survival for front line, advanced stage NSCLC patients from the INSIGHT interim analysis population. Kaplan-Meier plots of OS for NSCLC patients receiving platinum-based chemotherapy and immunotherapy (including platinum-based immunotherapy combinations). **5A)** OS compared between KRAS MUT versus KRAS WT patients treated with platinum-based chemotherapy **5B)** OS by VeriStrat classification in exclusively KRAS MUT patients treated with platinum-based chemotherapy **5C)** OS compared between KRAS MUT versus KRAS WT patients treated with immunotherapy **5D)** OS by VeriStrat classification in exclusively KRAS MUT patients treated with immunotherapy.

CONCLUSION

- Turnaround time for test results for the patients enrolled in the INSIGHT study was 34.7 hours.
- Presence of a detected KRAS mutation was a poor prognostic marker for front line, advanced stage NSCLC patients who received platinum-based chemotherapy, but not for patients who received immunotherapy.
- Combining blood-based proteomic (the VeriStrat® test) and genomic (the GeneStrat® test – KRAS) testing provides more complete information on a patient's prognosis and can help guide treatment decisions.

REFERENCES

1. Mellert, et al. J Mol Diagn. 2017, 19: 404-416.

RAS Targeted Drug Discovery

September 18, 2019 (Boston, MA)

Contact: yishan.chuang@biodesix.com