Abstract No: 765P

Comprehensive analysis of the association between RAS mutation and immune checkpoint marker expression

Hirotaka Miyashita[1]*, Razelle Kurzrock[2]*, Suzanna Lee[3], Sarabjot Pabla[4], Mary Nesline[4], Sean Glenn[4,5], Jeffrey M. Conroy[4], Paul DePietro[4], Shumei Kato[3]

1 Mount Sinai Beth Israel, Internal Medicine, NY, USA, 2 Worldwide Innovative Network (WIN) for Personalized Cancer Therapy, 3 Center for Personalized Cancer Therapy and Division of Hematology and Oncology, Department of Medicine, UC San Diego Moores Cancer Center, La Jolla, CA, USA, Department of Pathology, University of California San Diego, CA, USA, 4 OmniSeq Inc., Buffalo, NY, USA, 5 Roswell Park Comprehensive Cancer Center, Center for Personalized Medicine, Buffalo, NY, USA

BACKGROUND

- Sotorasib, *KRAS* G12C inhibiting small molecule, became the first to be approved by FDA in 2021 as a treatment targeting RAS. (1)
- Immune checkpoint blockade (ICB) has been incorporated to standard treatments for various types of cancer.
- There is in-vivo evidence suggesting the association between *KRAS* mutation and resistance to ICB. (2)
- In contrast, a retrospective study on the patients with advanced NSCLC showed no significant difference in ICB response depending on *KRAS* mutational status. (3)
- Revealing the relationship between RAS mutation and the expression level of the immune checkpoint molecules can guide potential combinations of treatment.
- We correlated RAS mutational status with mRNA expression levels of 16 immune checkpoint markers in a pan-cancer cohort.

METHODS

- We analyzed 16 immune checkpoint marker expression in 498 samples of patients with wide variety of cancer.
- 16 markers include: ADORA2A, BTLA, CD276, CTLA4, IDO1, IDO2, LAG3, NOS2, PD1, PDL1, PDL2, PVR, TIGIT, TIM3, VISTA and VTCN1.
- RNA expression was quantified by RNA sequence at OmniSeq laboratory.
- Transcript abundance was normalized to internal housekeeping gene profiles and ranked (0-100 percentile) to standardized by internal a reference population of 735 tumors spanning 35 histologies. The expression profiles were stratified by rank values into “Low” (0-24), “Intermediate” (25-74), and “High” (75-100).
- Genomic variants in the tumor samples and/or serum cell-free DNA (cfDNA) were analyzed with next-generation sequencing and the mutational status of the RAS gene family (*KRAS*, *NRAS*, and *HRAS*) were summarized.
- All investigations followed the guidelines of the UCSD Institutional Review Board for data collection (Study of Personalized Cancer Therapy to Determine Response and Toxicity, UCSD_PREDICT, NCT02478931) and for any investigational therapies for which the patients consented.

RESULTS

Figure. 1

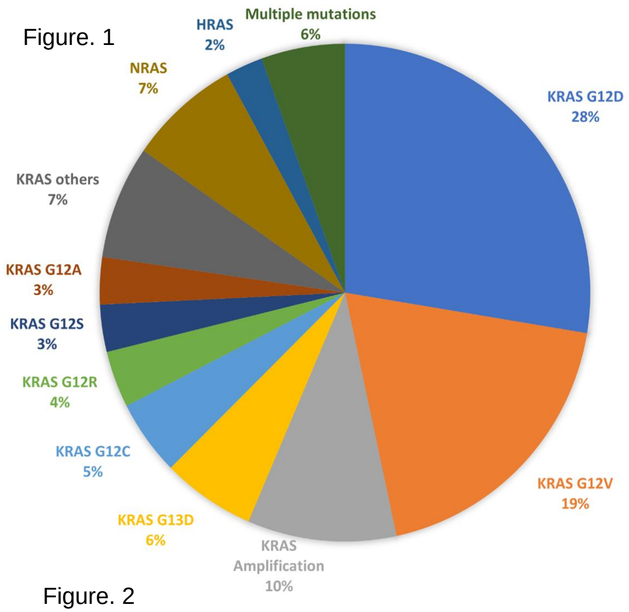

Figure. 2

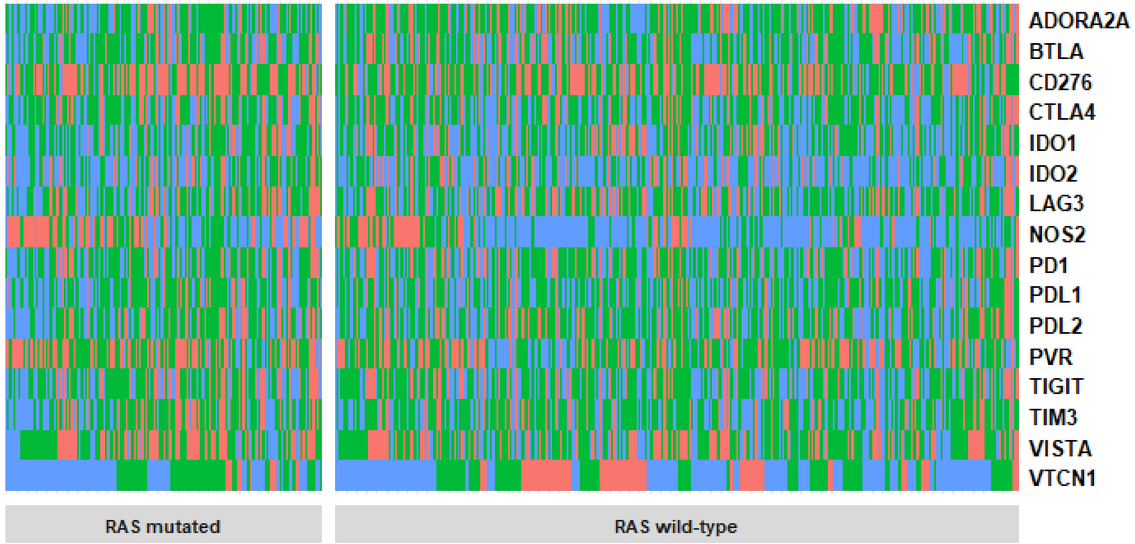

Figure 1. (Left) Characteristics of mutations in the patients harboring RAS mutation. (N = 163)

The most common mutation was *KRAS* G12D mutation followed by *KRAS* G12V and *KRAS* amplification. *NRAS* and *HRAS* mutations are seen in 7% and 2% of patients with any RAS mutation.

Figure 2. (bottom) Heatmap of immune checkpoint expression depending on RAS mutational status.

The color of blue, green and red means Low (rank value: 0-24), Intermediate (25-74) ,and High (75-100) expression.

Visually, high expression of NOS2 is more frequently seen in patients with RAS mutation. In contrast, frequency of high expression of VTCN1 is less frequent in patients with RAS mutation.

RESULTS

- The most common types of cancer in the cohort were colorectal and pancreatic cancer. (27.7% and 10.8%, respectively)
- RAS-mutant patients had a significantly larger proportion of high NOS2 expression compared to RAS-wild type patients (35.0% vs 17.0%, $p < 0.001$).
- In contrast, RAS-wild type patients had a significantly larger proportion of VTCN1 (B7-H4) high expression compared to RAS-mutant patients. (23.3% vs 8.0%, $p < 0.001$).
- After adjusting for tumor type, sex, and age, a significant inverse association between high VTCN1 expression and RAS-mutant status was seen ($p = 0.012$).
- No other checkpoint markers including PD-1, PD-L1, CTLA4 or LAG3, showed significant association with RAS status.

CONCLUSIONS

- RAS mutations were inversely associated with high expression of VTCN1 in this pan-cancer cohort.
- Although there is no medication to target VTCN1 and our data do not support the combination therapy of potential VTCN1 inhibition and RAS inhibition, the association between RAS status and VTCN1 expression suggests the relationship between RAS and immune checkpoint.
- Further study on the correlation between genetic mutation and immunological status of cancer may suggest a potentially effective combination therapy.

REFERENCES

1. Skoulidis F et al., Sotorasib for Lung Cancers with KRAS p.G12C Mutation, N Engl J Med. 2021 Jun, 24;384(25):2371-2381.
2. Yang B et al., MEK Inhibition Remodels the Immune Landscape of Mutant KRAS Tumors to Overcome Resistance to PARP and Immune Checkpoint Inhibitors, Cancer Res. 2021 May 15;81(10):2714-2729.
3. Jeanson A et al., Efficacy of Immune Checkpoint Inhibitors in KRAS-Mutant Non-Small Cell Lung Cancer (NSCLC), J Thorac Oncol. 2019 Jun;14(6):1095-1101.