MET gene copy number gains evaluated by NGS is more predictive than other methods to enrich for papillary RCC patients sensitive to Savolitinib, a selective MET inhibitor

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Background

There is no approved therapy for the treatment of papillary renal cell carcinoma (PRCC). Advances in molecular profiling of PRCC have identified a segment of PRCC with 10.6% MET mutation rates[1]. Chromosome 7 gains is a hallmark of PRCC and thought to occur at 50% frequency in PRCC[2]. We undertook a retrospective analysis of archival tumors to evaluate MET pathway aberrations for correlation with efficacy in a phase II study of Savolitinib (volitinib, AZD6094, HMPL-504) in patients with PRCC (NCT02127710). Archival diagnostic tumor samples were mandated for central confirmation of PRCC diagnosis, histological subtyping and for exploratory biomarker analysis.

Methods

84 archival tumors were obtained from the Ph2 Clinical Trial and profiled using four methods: H&E stain for PRCC histological subtype, immunohistochemistry (IHC) for c-Met protein expression (Ventana, CONFIRM), FISH for MET gene amplification (Abbott, VYSIS), Next Generation Sequencing (NGS) as an orthogonal method for confirmation of MET amplifications, detection of HGF gene amplifications, MET mutations, chromosome 7 ploidy, and other exploratory genomic biomarkers (Foundation Medicine Inc,T7 panel).

Biomarker Hypotheses

Biomarker	PRCC Frequency	Central test	Protocol endpoint
PRCC histological subtype [3]	Type I: 47% Type II: 37% RCC Unclassified: 16%	**H&E**	Mandatory
Chromosome 7 gains [2]	43-75%	**NGS**	Exploratory
MET gene amplification [3]	46% of Type II 81% of Type I	**FISH or NGS**	Exploratory
Met over-expression	n/a	**IHC**	Exploratory
MET gene mutations [1] 400 gene panel	5-13%	**NGS**	Exploratory

Table 1. Predictive Biomarker Hypotheses. Central tests for all submitted and evaluable archival tumor tissues at onset of trial.

Savolitinib confers efficacy in a subgroup of PRCC patients

Tumors from 8 patients who had a confirmed partial response (PR) to Savolitinib were used to identify the most predictive biomarker test. Retrospective analysis demonstrated that only NGS, not IHC, FISH or histological subtype, identified all Savolitinib PRs.

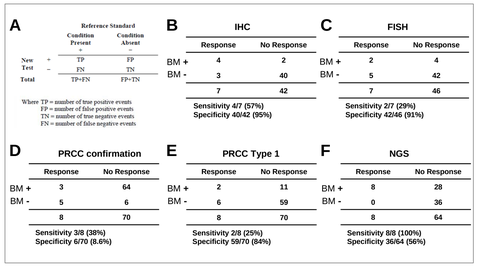


Figure 1. Sensitivity and Specificity of biomarker (BM) methods to identify most predictive test.
A) CDRH reference **B)** MET IHC by H-Score ≥ 200 **C)** MET FISH by average MET gene copy number ≥ 4 **D)** PRCC central histological confirmation by H&E and pathology **E)** PRCC histological subtype 1 only by H&E and central pathology **F)** NGS by MET kinase domain mutation and MET gene copy number gain (focal ≥ 6 or chromosomal ≥ 3).
Sensitivity is how often the test is positive when response is present (TP /TP+FN)
Specificity is how often the test is negative when response is absent (TN /FP+TN)

Assessment of the MET Pathway by NGS

NGS targeted ~400 gene panel (version T7; Foundation Medicine Inc, Cambridge, MA) of archival tumor was used as previously described[5]. PRCC was confirmed as **"MET driven"** by identification of MET copy number gain (chromosome 7 gain or focal MET amplification ≥ 6 copies), HGF gene amplification (≥ 6 copies), or MET kinase domain mutations (allele frequency>5%)[4].

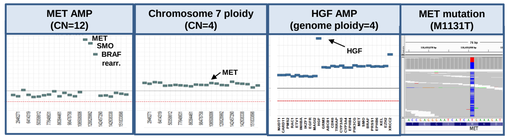
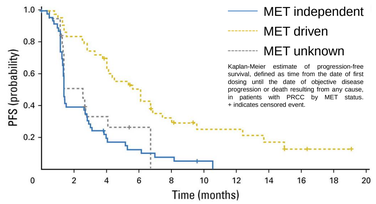


Figure 2. Savolitinib Responder Patient Tumor NGS profiles. Chromosome 7 profile (three left hand site panels) from 27 genes on Chromosome 7 in NGS test + 4200 SNPs with bioinformatic analytics to determine genome ploidy. Integrated Genomic Viewer highlights the single nucleotide variant in the kinase domain of the *MET* gene (right hand panel).



Time, months	0	1.4	2.8	4.1	5.5	6.9	8.3	9.7	11.1	12.5	13.9	15.2	16.6	18.0	19.4
No. at risk:															
MET independent	46	30	15	8	4	3	2	1	0						
MET driven	44	39	29	20	13	9	6	6	4	3	3	3	0		
MET unknown	19	13	3	2	1	0									

Figure 3. Kaplan-Meier estimate of PFS by MET-driven NGS biomarker status. Adapted from Ph2 trial publication with original legend (inset) [4]

Frequency of MET-driven PRCC NGS biomarkers

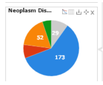
Biomarker	Frequency			
	TCGA [1] (n=157, Tx naïve)		NCT02127710 [4] (n=92**, Ph2 trial)	
Chromosome 7 gain*	62/157	39.5%	29/92	31.5%
MET amplification	2/157	1%	9/92	10%
MET kinase domain mutation	10/157	6%	7/92	7.5%
HGF amplification	0/157	0%	1/92	1%
Total	**74/157**	**46.5%**	**46/92**	**50%**

(*) where Chromosome 7 gain is the only event
(**) 8 cases profiled with no associated RECIST data

Table 2. Frequency of MET-driven Biomarkers in Ph2 Trial Compared to The Cancer Genome Atlas (TCGA) Published Cohort. TCGA and Ph2 trial do not use the same NGS platform and therefore direct comparison is only observational. An instance of TCGA sequencing data was used to obtain raw NGS data to which the same bioinformatic pipeline (AZ internal) was used in both PRCC cohorts to describe chromosome 7 changes.

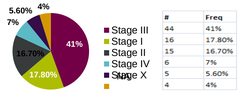
Disease Stage, NGS Biomarker and Response

TCGA cohort tends to be early stage disease



Neoplasm Dise...	#	Freq
Stage I	173	59.2%
Stage III	52	17.8%
NA	29	9.9%
Stage II	23	7.9%
Stage IV	15	5.1%

Ph2 trial population has spectrum of stages



	#	Freq
Stage III	44	41%
Stage I	16	17.80%
Stage II	15	16.70%
Stage IV	6	7%
Stage X	5	5.60%
NA	4	4%

Figure 4. Disease Stage in Ph2 Trial and TCGA PRCC patient populations. Comparisons are only observational since disease stage proportions are different.

MET-Driven NGS biomarkers and PRCC disease association

Biomarker	Histology (central)			
	Type 1	Type 2	Unclassified	Other (not PRCC)
Chromosome 7 gain*	4 (1PR)	23 (1PR)	4 (2PR)	2
MET amplification	4 (1PR)	2 (1PR)	1PR	0
MET kinase mutation	3	3	0	0
HGF amplification	0	0	1PR	0
NOT MET-driven	0	27	5	0
Total**	**11**	**55**	**11**	**2**

(*) where Chromosome 7 gain is the only event
(**) 5 patients where histology not available at time of analysis

Table 3. Redefining the histological classification of PRCC. MET-driven PRCC using NGS biomarkers is not only associated with Type 1 PRCC. Further, MET kinase mutations are identified in both Type 1 (as expected from the literature) and Type 2 PRCC. PR=partial responders n=8 in the Ph2 Savolitinib trial. These two observations demonstrate the MET-driven PRCC is not only contained in the histological Type 1 subgroup of PRCC.

Mutual Exclusivity / Overlap of MET-driven NGS Biomarkers

Venn Diagram is not to scale

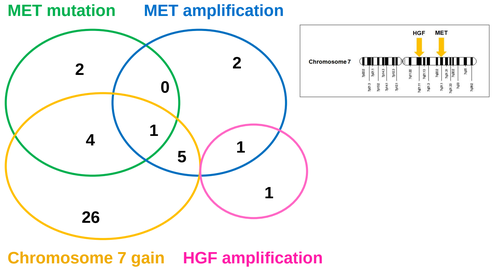


Figure 5. MET-driven PRCC by NGS biomarker category in Savolitinib Ph2 Trial. MET and HGF loci are both located on Chromosomes 7 (inset). Although each biomarker can exist independently, some PRCC cases have co-occurence of MET-driven NGS biomarkers.

Conclusions

- NGS is the most predictive test for response to Savolitinib.
- The frequency of MET Driven NGS biomarkers in the Ph2 population were 50% and captured all 8 responders, however 42% of non-responders are also MET-driven.
- Molecular characterization of MET status by NGS was more predictive of response to Savolitinib than a classification based on pathology, redefining PRCC at the molecular level and identifying MET as a target in PRCC.
- Biomarker driven patient selection should be considered for targeted therapies to the MET Receptor Tyrosine Kinase in PRCC and may apply to other indications.

References
1. Linehan et al. NEJM 2016
2. Jiang et al. Am J Pathol 1998
3. Albiges et al. CCR 2014
4. Choueiri et al. JCO 2017
5. Frampton et al. Nat Biotech 2013

Acknowledgements
Thank you to the patients who participated in savolitinib clinical trials and to their families.

Supported by AstraZeneca

Presented at the AACR annual meeting 2018, April 14–18th in Chicago IL,USA