Poster board #48B

Proteomic analysis of MET tyrosine kinase receptor as a prognostic biomarker of survival in Patients with Gastroesophageal Cancer

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Background

- Overexpression of MET in gastroesophageal cancer (GEC) is associated with poor prognosis and potentially predictive of benefit from anti-MET therapies.
- We developed a clinically-validated multiplex MS assay to measure MET protein level in FFPE GEC tissue.
- Archival tumor specimens were collected in the United States and Italy and included full clinical annotations (staging, HER2 status, treatment, and overall survival)
- We are running the assay in a CLIA-certified-CAP-accredited laboratory to concurrently assess protein expression levels for MET and other diagnostic and potentially targetable biomarkers, e.g. EGFR, HER2, HER3, FGFRs, PDL1 and IGF1R.

Material & Methods

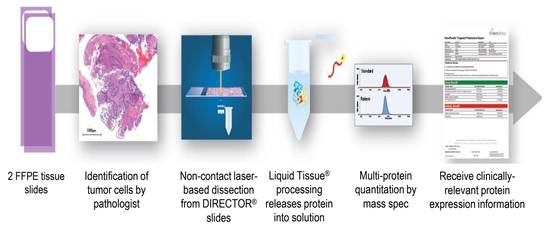


Figure 1. Liquid Tissue®-SRM workflow for analysis of proteins from FFPE tissue

Analytical Performance of MET Assay

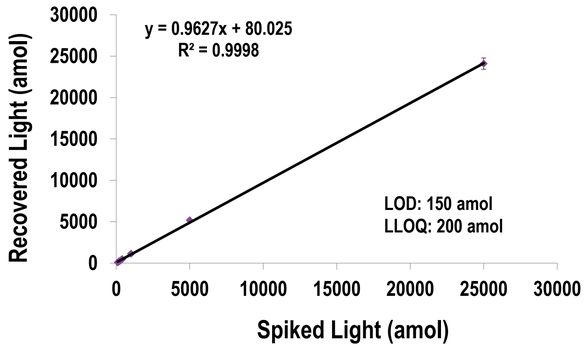


Figure 2. Representative standard curve for MET in eukaryotic matrix. The calibration curve was built by adding various amount of unlabeled (light) synthetic MET peptide into a matrix obtained from formalin-fixed SKBR3 cells containing 5 fmol of isotopically-labeled MET peptide.

Results

SRM Measurement is Highly Concordant with *MET* Copy Number or *MET/CEP7* Ratio

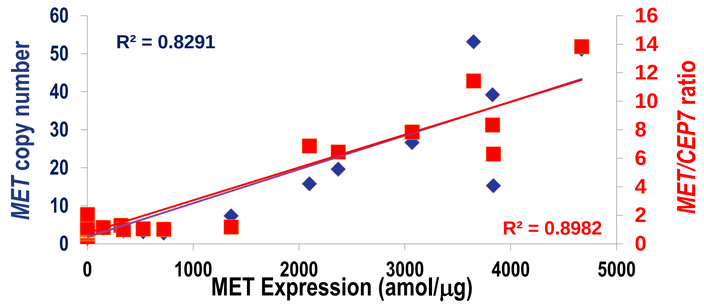
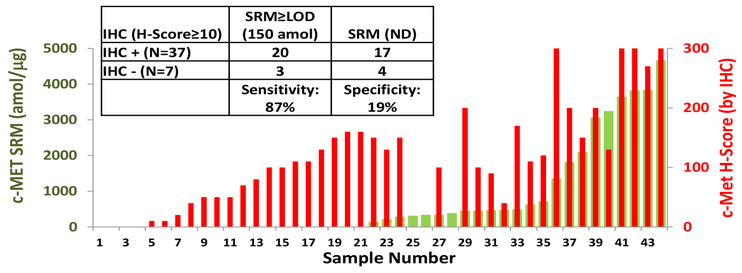


Figure 3. Comparison of MET protein level and MET gene copy number in GEC tissues (N=30). The MET SRM result is plotted against *MET* GCN (blue) or *MET:CEP7* ratio (Red).

Poor correlation of MET IHC with targeted proteomics in GEC tumors



IHC (H-Score≥10)	SRM≥LOD (150 amol)	SRM (ND)
IHC + (N=37)	20	17
IHC - (N=7)	3	4
	Sensitivity: 87%	Specificity: 19%

Kaplan-Meier Plot of OS by MET Status (cutoff of 400 amol/µg)

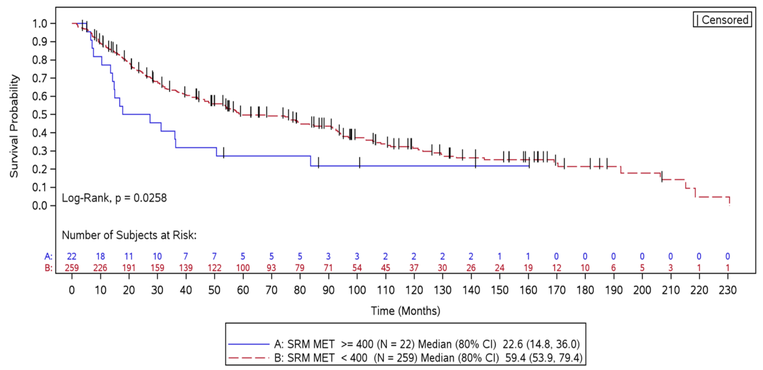


Figure 4. Median survival at the 400amol/ug cutoff Met-SRM value was 22.6 months for Met-positive patients vs 59.4 months for Met-negative patients

Quantitation of MET in clinical FFPE GEC tumors

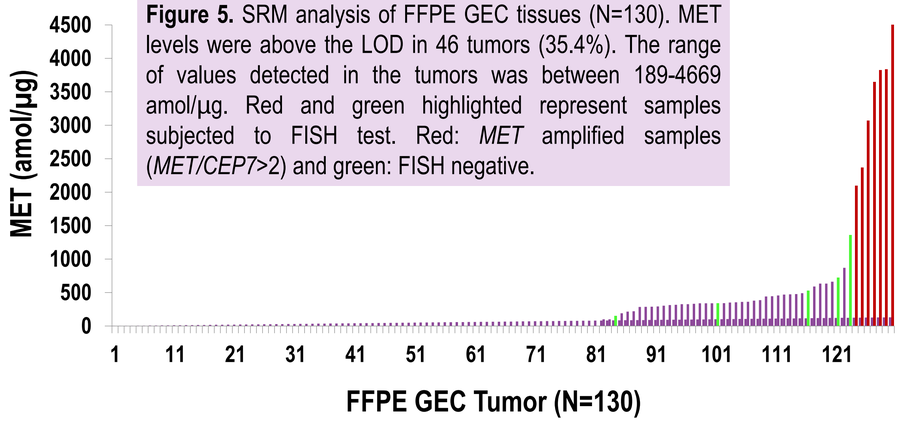


Figure 5. SRM analysis of FFPE GEC tissues (N=130). MET levels were above the LOD in 46 tumors (35.4%). The range of values detected in the tumors was between 189-4669 amol/µg. Red and green highlighted represent samples subjected to FISH test. Red: *MET* amplified samples (*MET/CEP7*>2) and green: FISH negative.

SRM analysis of FFPE GEC tumors (N=130)			
	MET SRM (amol/µg)		
	ND (<LOD)	150-1500	>1500
Sample #	84	39	7
Percentage	64.6%	30%	5.4%

SRM vs. FISH (N=30)	MET SRM (amol/µg)		
	ND	150-1500	>1500
Sample #	17	6	7
MET amplified (*MET/CEP7*≥2)	0	0	7
Percentage positive	0%	0%	100%

Table 1: Summary for MET expression in GEC tumors (N=130). Upper table shows that 5.4% of GEC have MET level >1500 amol (7/130). Using this value (>1500 amol/mg) as the cut off, the MET assay reliably detected *MET* amplified GEC tumors with 100% sensitivity and 100% specificity (shown in lower table).

Conclusions

- We have developed a mass spectrometry-based assay to measure the absolute level of MET in clinical FFPE tumor tissues with high level of specificity and temporal stability, and quick turn around time (5 days from time of tissue receiving).
- The SRM assay is able to detect *MET* amplified samples with high sensitivity and specificity as compared to FISH.
- *MET* amplification by FISH and MET expression by SRM were independent prognostic biomarkers. Compared with IHC, SRM may provide superior benefit towards informed decisions about MET-targeted therapy.
- The ability to concurrently quantify MET and other relevant proteins represents a novel clinical tool for efficient tumor expression profiling, leading to better informed therapeutic decisions for patients with GEC.