Evaluating CSF circulating tumor DNA in intraparenchymal brain metastasis

Stephanie Cheok, MD[1*], Azeet Narayan, PhD[2], Anna Arnal-Estape, PhD[3,5], Scott Gettinger, MD[4,5], Sarah B. Goldberg, MD, MPH[4,5], Harriet M. Kluger, MD[4,5], Don Nguyen, PhD[3,4,5 ◊], Abhijit Patel, MD, PhD[2,5], Veronica Chiang, MD[1,5]

Introduction

Discordant responses between brain and systemic metastases are not infrequently seen in patients receiving targeted therapies. In these cases, repeat tumor profiling of the progressing site could guide further therapy. We propose that circulating tumor DNA (ctDNA) might be detectable in cerebrospinal fluid (CSF) and reflective of the genetic profile of treatment-resistant intraparenchymal brain metastases.

Patients and Methods

Patients with progressive brain metastases undergoing a craniotomy or lumbar puncture were enrolled between July 2018 to April 2019 under an IRB-approved protocol. CSF and blood were collected simultaneously. Cell-free DNA (cfDNA) was extracted using a QIAamp MinElute Virus Vacuum Kit (Qiagen). Tumor-derived somatic mutations (ctDNA) were identified and quantified using an Error-Suppressed Deep Sequencing method previously published by our group. Forty-three mutation-prone regions of common 24 cancer-associated genes were assayed, and the allelic fractions were calculated against wild-type sequence counts.

CHARACTERISTIC	N = 16 (%)
AGE	
mean ± SD	65.04 ± 12
median	61
GENDER	
Female	6 (37.5%)
Male	10 (62.5%)
CANCER PRIMARY	N=14/16
Lung	10 (71.4%)
NSCLC	7
SCLC	2
LCLC	1
Melanoma	2 (0.95%)
Breast	1 (0.71%)
Renal	1 (0.71%)
Colorectal	1 (0.71%)
NPH	2
CSF SOURCE	
Craniotomy	13 (54.52%)
LP/LD	10 (43.48%)

Table 1: Patient Demographics and disease distribution

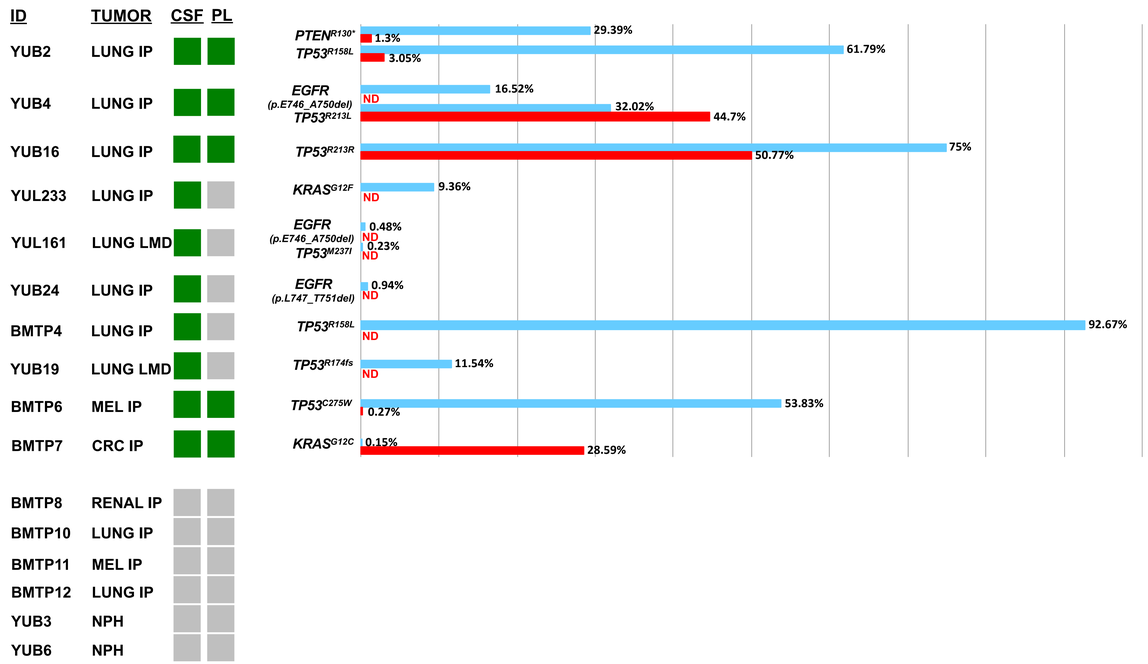

Figure 1: Quantification of cfDNA mutations in time-matched CSF and plasma of patients with brain metastasis. The amount of mutated DNA within the total cell-free DNA is shown as the mutant allele fraction (% AF).

Results

Sixteen patients were enrolled in this study - 12 patients with intraparenchymal brain metastases, 2 patients with CSF cytology-positive leptomeningeal disease (LMD) and 2 patients with normal pressure hydrocephalus (NPH) as controls. (Table 1) Primary cancer types were lung (n=10), melanoma (n=2), renal cell (n=1) and colorectal (n=1) cancers. cfDNA was found in all sixteen samples of CSF. CSF ctDNA was found in 8 patients (67%) and plasma ctDNA was only found in 5 patients (42%) with intraparenchymal lesions. CSF ctDNA but not plasma ctDNA was detected in both LMD patients. No ctDNA was identified in the CSF or blood of either NPH patient. Six patients in the cohort also had time-matched brain metastasis tissue. In 4 of 6 patients (67%), congruent ctDNA mutations were found in the CSF and tissue while congruent ctDNA was only found in one out of six plasma samples (17%).

Discussion and Conclusion

Analysis of CSF can be a viable alternative to obtaining brain metastasis tissue for DNA profiling in the detection of brain metastasis resistance mutations. The presence CSF ctDNA is not restricted to LMD and was isolated from two-thirds of patients with intraparenchymal disease in our cohort. Furthermore, CSF remains a better source than plasma for the detection of ctDNA across multiple brain metastases tumor subtypes.

Contact

Stephanie Cheok
Yale Department of Neurosurgery
stephanie.cheok@yale.edu

Affiliations

[1]Department of Neurosurgery, Yale University, New Haven, Connecticut, USA
[2]Department of Therapeutic Radiology, Yale University, New Haven, Connecticut, USA
[3]Department of Pathology, Yale University, New Haven, Connecticut, USA
[4]Department of Medicine (Medical Oncology), Yale University, New Haven, Connecticut, USA
[5]Yale Cancer Center, New Haven, Connecticut, USA