Chemotherapy: Open Access

Davis, et al., Chemo Open Access 2017, 6:2
DOI: 10.4172/2167-7700.1000230

Short Communication OMICS International

Is Circulating Tumor DNA (Ctdna) Use Ready For Prime Time? Applications and Challenges of Ctdna in the Era of Precision Oncology

Davis AA, Chae YK* and Giles FJ

Department of Medicine, Division of Hematology Oncology, Feinberg School of Medicine, Northwestern University, Chicago, IL 60611, USA

***Corresponding author:** Young Kwang Chae, Department of Medicine, Division of Hematology Oncology, Feinberg School of Medicine, Northwestern University, Chicago, IL 60611, USA, Tel: 312 926 4248; Fax: 312 695 0370; E-mail: young.chae@northwestern.edu

Received date: March 26, 2017; **Accepted date:** April 17, 2017; **Published date:** April 22, 2017

Introduction

"Liquid biopsies" have emerged as a tool to monitor genomic alterations in the peripheral blood [1]. Cell-free DNA consists of non-cancerous nucleic acids and circulating tumor DNA (ctDNA). The proportion of ctDNA depends on the tumor cell of origin and stage of malignancy [2-5]. Peripheral blood biopsies can detect single nucleotide variants, indels, copy number variants, rearrangements and fusions, non-invasively, avoiding risk associated with repeat tissue biopsies. However, concerns remain with respect to ctDNA adequately reflecting tumor heterogeneity, thresholds for detection, and lack of randomized control trials to validate improved survival outcomes. As a result, further data are necessary to compare sequencing data between tissue and blood biopsies to validate clinical utility.

Our recently published paper entitled "Concordance between genomic alterations assessed by next-generation sequencing (NGS) in tumor tissue or circulating cell-free DNA" compared mutational profiles across two commercial NGS platforms [6]. NGS is high-throughput sequencing that involves processing millions of DNA fragments. The ctDNA platform in our study utilized massively parallel and deep digital sequencing from a blood sample [7]. In contrast, tissue NGS involved parallel DNA sequencing from formalin-fixed or paraffin-embedded specimens. We utilized clinical reports from paired tissue processed via FoundationOne and blood samples processed Guardant 360. FoundationOne was a 315 gene panel, while the blood biopsy examined 68 genes. We compared the 65 genes that overlapped between both assays. Our study was a retrospective analysis of 28 patients with a variety of advanced solid tumors. We compared concordance at the gene variant level, meaning that only genomic alterations with the exact same nucleotide sequence were considered concordant. We also performed sensitivity, specificity, and diagnostic accuracy analyses. The goal of our study was to systematically examine variant-level concordance across a diverse group of advanced solid tumors.

Contrary to prior reports in the literature that have reported high concordance for particular driver mutations, our study found that concordance when genomic alterations were detected using either biopsy ranged from 11.8-17.1% [8,9]. There are several reasons to explain the differences in reported concordance values. First, our lower concordance analyses excluded wild type/wild type variants (e.g., when no mutation was detected in the same gene in both studies). Second, we examined a larger sequencing region and restricted our definition of concordance to only include the exact same genomic alteration down to the nucleic acid level. This was critical in order to determine whether these NGS platforms could reliably detect specific resistance mutations, such as *EGFR* T790M in lung cancer. Recently, Kuderer and colleagues compared two commercially available NGS platforms validated our work in a sample of nine patients with heterogeneous histologies [10]. In this study, the authors reported a concordance rate of 22% for detected genomic alterations when comparing the same two commercial assays. The study further reported that only 25% of drugs were recommended for the same patient by both platforms. Clearly, our study and the report by Kuderer and colleagues demonstrate a high level of discordance that warrants further investigation.

Another important finding of our study was that while more mutations were detected in tissue biopsies as compared to ctDNA, both tissue and ctDNA contributed unique information about the genomic profile of a patient's tumor. Specifically, our study found that over half of mutations detected using one NGS technique were not detected using the other method. A primary reason for this discordance is likely the inherent differences in the biopsy techniques. While ctDNA biopsies rely on detecting tumor DNA that is released into the blood via apoptosis or necrosis, tissue biopsies only target a single primary or metastatic lesion [11]. Technical differences are also important with respect to possible false positives in ctDNA at low variant allele frequency. In fact, a recent study reported that approximately 11% of healthy controls were found to have a genomic alteration in *TP53* in the peripheral blood [12]. There are several additional challenges with respect to detecting ctDNA in the blood. First, it is critical to carefully evaluate ctDNA assays in order to distinguish cfDNA from true ctDNA. Second, the half-life of ctDNA is short (<1.5 hours), which suggests that ctDNA is a highly dynamic marker influenced by tumor shedding [13]. Finally, ctDNA is being explored for early detection of cancer, but additional studies are necessary to determine the sensitivity of these assays in control populations [14].

A clear advantage of liquid biopsies is the accessibility of obtaining samples from patients, enabling serial biopsies to reflect the evolving genomic profile of an individual's tumor. A recent study demonstrated that serial testing for *TP53* mutations in patients with ovarian cancer using ctDNA may be more specific for disease monitoring as compared to serum CA-125 [15]. This potential exists because *TP53* genomic alterations are present in approximately 99% of patients with high-grade serous ovarian carcinoma. Interestingly, the mutant allele frequency of ctDNA was more prognostic than CT imaging, a finding that was previously validated in breast cancer that ctDNA was an earlier marker for disease relapse than CT imaging [16]. In addition, ctDNA has been shown to reflect minimal residual disease and recurrence in stage II colorectal cancer and to have prognostic value in non-small cell lung cancer (NSCLC) [17,18].

We are currently pursuing additional studies to compare these two platforms further. While our initial study compared sequencing data across a heterogeneous group of solid tumors, we are currently exploring how concordance may vary by tumor histology, as previous data have demonstrated that certain tumors are more (lung or breast

cancer) or less (glioblastoma multiforme) likely to be captured in peripheral blood [2]. Second, we are comparing summative measures of genomic instability (e.g., tumor mutational burden (TMB)) across tissue and liquid biopsies. Previous research has indicated potential for TMB to predict response to immunotherapy using NGS tissue biopsies [19,20]. It remains to be seen whether shorter regions of the genome in the peripheral blood may predict similar response to immune checkpoint blockade.

We envision that ctDNA may have clinical utility for serial assessments to identify particular resistance mutations given the high specificity of the assay (e.g., *EGFR* T790M in NSCLC and *ESR1* in breast cancer) and to identify disease relapse or progression prior to radiologic evidence of disease. Certainly, cost of repeat peripheral blood biopsies remains an issue. However, cost effectiveness would be promoted by matching patients with an ideal treatment and then switching therapy as the tumor evolves to save costs associated with unnecessary treatment, side effects, and imaging.

Collectively, these studies highlight the complex biology of advanced solid tumors. There are many challenges that exist with regard to spatial and temporal tumor heterogeneity, accessibility of repeat tissue sampling, ability to detect ctDNA at very low detection thresholds, and inherent differences in sequencing and sampling techniques between the biopsies. For these reasons, we envision a complementary approach using both tissue and ctDNA in the near future to optimally capture and monitor an evolving mutational profile when cost becomes less prohibitive.

References:

1. Alix-Panabieres C, Pantel K (2016) Clinical Applications of Circulating Tumor Cells and Circulating Tumor DNA as Liquid Biopsy. Cancer Discov 6: 479-491.
2. Bettegowda C, Sausen M, Leary RJ, Kinde I, Wang Y, et al. (2014) Detection of circulating tumor DNA in early- and late-stage human malignancies. Sci Transl Med 6: 224ra24.
3. Forshew T, Murtaza M, Parkinson C, Gale D, Tsui DW, et al. (2012) Noninvasive identification and monitoring of cancer mutations by targeted deep sequencing of plasma DNA. Sci Transl Med 4: 136ra68.
4. Beaver JA, Jelovac D, Balukrishna S, Cochran RL, Croessmann S, et al. (2014) Detection of cancer DNA in plasma of patients with early-stage breast cancer. Clin Cancer Res 20: 2643-2650.
5. Couraud S, Vaca-Paniagua F, Villar S, Oliver J, Schuster T, et al. (2014) Noninvasive diagnosis of actionable mutations by deep sequencing of circulating free DNA in lung cancer from never-smokers: a proof-of-concept study from BioCAST/IFCT-1002. Clin Cancer Res 20: 4613-4624.
6. Chae YK, Davis AA, Carneiro BA, Chandra S, Mohindra N, et al. (2016) Concordance between genomic alterations assessed by next-generation sequencing in tumor tissue or circulating cell-free DNA. Oncotarget 7: 65364-65373.
7. Lanman RB, Mortimer SA, Zill OA, Sebisanovic D, Lopez R, et al. (2015) Analytical and Clinical Validation of a Digital Sequencing Panel for Quantitative, Highly Accurate Evaluation of Cell-Free Circulating Tumor DNA. PLoS One 10: e0140712.
8. Schwaederle M, Husain H, Fanta PT, Piccioni DE, Kesari S, et al. (2016) Use of Liquid Biopsies in Clinical Oncology: Pilot Experience in 168 Patients. Clin Cancer Res 22: 5497-5505.
9. Zill OA, Greene C, Sebisanovic D, Siew LM, Leng J, et al. (2015) Cell-Free DNA Next-Generation Sequencing in Pancreatobiliary Carcinomas. Cancer Discov 5: 1040-1048.
10. Kuderer NM, Burton KA, Blau S, Rose AL, Parker S, et al. (2016) Comparison of 2 Commercially Available Next-Generation Sequencing Platforms in Oncology. JAMA Oncol.
11. Jahr S, Hentze H, Englisch S, Hardt D, Fackelmayer FO, et al. (2001) DNA fragments in the blood plasma of cancer patients: quantitations and evidence for their origin from apoptotic and necrotic cells. Cancer Res 61: 1659-1665.
12. Fernandez-Cuesta L, Perdomo S, Avogbe PH, Leblay N, Delhomme TM, et al. (2016) Identification of Circulating Tumor DNA for the Early Detection of Small-cell Lung Cancer. EBioMedicine 10: 117-123.
13. Diehl F, Schmidt K, Choti MA, Romans K, Goodman S, et al. (2008) Circulating mutant DNA to assess tumor dynamics. Nat Med 14: 985-990.
14. Siravegna G, Marsoni S, Siena S, Bardelli A (2017) Integrating liquid biopsies into the management of cancer. Nat Rev Clin Oncol.
15. Parkinson CA, Gale D, Piskorz AM, Biggs H, Hodgkin C, et al. (2016) Exploratory Analysis of TP53 Mutations in Circulating Tumour DNA as Biomarkers of Treatment Response for Patients with Relapsed High-Grade Serous Ovarian Carcinoma: A Retrospective Study. PLoS Med 13: e1002198.
16. Dawson SJ, Tsui DW, Murtaza M, Biggs H, Rueda O, et al. (2013) Analysis of circulating tumor DNA to monitor metastatic breast cancer. N Engl J Med 368: 1199-1209.
17. Tie J, Wang Y, Tomasetti C, Li L, Springer S, et al. (2016) Circulating tumor DNA analysis detects minimal residual disease and predicts recurrence in patients with stage II colon cancer. Sci Transl Med 8: 346ra92.
18. Pecuchet N, Zonta E, Didelot A, Combe P, Thibault C, et al. (2016) Base-Position Error Rate Analysis of Next-Generation Sequencing Applied to Circulating Tumor DNA in Non-Small Cell Lung Cancer: A Prospective Study. PLoS Med 13: e1002199.
19. Rizvi NA, Hellmann MD, Snyder A, Kvistborg P, Makarov V, et al. (2015) Cancer immunology. Mutational landscape determines sensitivity to PD-1 blockade in non-small cell lung cancer. Science 348: 124-128.
20. Snyder A, Makarov V, Merghoub T, Yuan J, Zaretsky JM, et al. (2014) Genetic basis for clinical response to CTLA-4 blockade in melanoma. N Engl J Med 371: 2189-2199.