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"Liquid Gold" - The unTAPped Potential of Cerebrospinal Fluid Analysis?

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SUMMARY

Obtaining blood and cerebrospinal fluid is generally less invasive than standard tumor biopsy, and are increasingly used to develop surrogate biomarkers. Leptomeningeal disease, a devastating complication of cancer, represents a unique opportunity for using liquid biopsies for diagnosis, treatment, and to elucidate underlying mechanisms of resistance to therapy.

See related article by Smalley et al., p. 2163

In this issue of *Clinical Cancer Research*, Smalley and colleagues analyzed serial cerebrospinal fluid (CSF) samples from patients with metastatic melanoma, some with confirmed leptomeningeal disease (1). Interestingly, the investigators incubated melanoma cells in CSF and analyzed its impact on gene expression, as well as therapeutic response to BRAF inhibitors. Proteomic/transcriptional signatures described an enrichment of pathways involved in innate immunity, IGF-related signaling, and protease-mediated damage, resulting also in relative BRAF resistance via the PI3K/AKT pathway.

This research is critical in understanding the central nervous system (CNS) and its own tumor microenvironment, as the incidence of metastases to the CNS is increasing with devastating impact (2). While progress is slowly but surely being made in the treatment of parenchymal brain metastases, involvement of the leptomeninges remains the most challenging and intractable complication of cancer. Leptomeningeal disease is identified in about 10% of patients with metastatic melanoma and has a dismal prognosis with survival counted in weeks to months, at best. Other solid tumors like breast and lung cancer have a similar incidence rate, resulting overall in a substantial patient population without meaningful treatment options (2).

Diagnosis of leptomeningeal disease remains challenging, especially as both radiologic and cytologic assessment can be equivocal. Presence of cancer cells in the CSF remains the gold standard but it has a low sensitivity, being positive in only about 50% of patients during their first CSF assessment via lumbar puncture. Modern approaches, including the analysis of circulating tumor cell analysis and cell-free tumor DNA sequencing (ctDNA), have allowed for improved diagnostic sensitivity of leptomeningeal disease in the research setting. However, it should be noted that parenchymal brain metastases that are in close proximity to the leptomeningeal space or ventricular system can also shed ctDNA into the CSF, potentially impacting the use of ctDNA in the diagnosis of leptomeningeal disease, with one example being pediatric tumors (3).

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While no large prospective clinical trial has been completed assessing the potential of longitudinal CSF and blood sampling in the pediatric tumor population, including primary CNS tumors, smaller studies have shown the feasibility of ctDNA detection across different pediatric cancers. Plasma samples from patients with neuroblastoma detected additional genomic alterations in the ctDNA that were not found in the primary tumor, and methylation analysis of other CNS tumors via liquid biopsy was superior to standard histopathologic diagnosis. In patients with solid tumors and brain metastases, differences between mutation patterns within the primary tumor, brain metastases, CSF, and plasma have been found. For example, a recent study evaluating the molecular characteristics of brain metastases of patients with non–small cell lung cancer found that the detection rate of EGFR mutations in CSF ctDNA was double compared with plasma, as well as an even higher detection rate in patients with leptomeningeal disease (4).

Furthermore, CSF and plasma are now being investigated for their potential value in clinical monitoring of therapeutic efficacy, recurrence, as well as the identification of mechanisms of resistance.

The contribution of this study to the field is quite significant as it highlights several key issues in investigating leptomeningeal disease. The fact that there was only 8 patients with serial samples, and this still representing "one of the largest cohorts" makes a strong case for the lack of data in this space, and the absolute need for improved and targeted investigation. The longitudinal sample collection allows to eliminate some degree of variability but one has to acknowledge the complex biology and the inherent significant variability of the clinical phenotypes represented in a small number of patients, including one long-term survivor. While this patient served as a control in this analysis, concerted efforts should be undertaken to see if other long-term survivors identified at other institutions have comparable CSF findings. In addition, this underlines the generally dismal outcomes as one of the main obstacles in prospective leptomeningeal disease research, significantly limiting our ability to meaningfully and statistically compare responders versus nonresponders. For clinical trial enrollment, washout periods from prior therapies need to be considered in light of short life expectancy, and translational research plans need to take into account a much shorter opportunity for longitudinal sampling and monitoring, making the identification of mechanism of response, let alone resistance exceptionally difficult.

The focus on proteomics in this study is very useful to better understand the microenvironment induced by CSF, where still very little data exist. The brain parenchymal and the leptomeningeal space represent microenvironments that are distinctly different from each other, but also different from the rest of the body, as certain cells are found only in the brain parenchyma or meninges, shielding the brain

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from hydrophilic solutes via the blood–CSF and blood–brain barrier (5). In addition to key differences in regards to this microenvironment, significant variances exist in regards to metabolic level and micronutrients and oxygen, as well as general immune cell composition. For example, the CSF is low not only in oxygen but also in metabolites, representing a substantial survival challenge for cancer cells which are typically very metabolically active, but somehow have adapted to this environment, and able to rapidly grow within this space.

Additional studies have suggested that melanoma brain metastases might have distinct molecular features, including an increased activation of the PI3K-protein kinase B (PI3K-AKT) signaling pathway, which has been implicated with resistance to BRAF and MEK inhibitors both in the preclinical and clinical setting. Using CSF for early recognition of the development of resistance to targeted therapy might be one of several future uses for liquid biopsies in general clinical practice. Furthermore, it has been shown that BRAF inhibitors can reach the CSF, but generally at lower levels and with significant patient-to-patient variability, supporting the need for more in depth analysis of drugs' ability to cross the blood–brain barrier (2). This is also particularly important, as the phenomenon of opposing therapeutic responses has been observed during the systemic treatment of intracranial and extracranial lesions in patients with melanoma with brain metastases. While intracranial response rates reported were equivalent to response rates for extracranial sites, the duration of response in the CNS was reduced by half (2). It remains unclear whether this is related to decreased drug exposure or to distinct resistance mechanism intracranially. This study indicates an element of resistance conferred by the microenvironment.

While the CNS used to be considered an immune cell poor environment, recent research has shown that blood circulating immune cells can enter the CNS, especially in the setting of CNS involvement with cancer. The investigators focused only on cell-free CSF, appropriately for proteomic analyses, however there is much to learn from the cellular component. As mentioned earlier, cancer cells must undergo the biggest adaptation for continued growth and spread if they colonize the leptomeningeal space. In addition, a deeper understanding of the immune response in the brain and CSF is direly needed, and a closer analysis of the cellular component is incredibly useful toward that end. With the efforts of ongoing clinical trials, including ones investigating the efficacy of either intravenous (NCT03091478 and NCT02939300) or intrathecal checkpoint inhibitors (NCT00338377) as well as intrathecal tumor-infiltrating lymphocytes (NCT00338377) in this patient group, serial liquid biopsies will hopefully offer deeper insights into the CSF as its own microenvironment.

Much remains unknown about the CSF microenvironment in patients with leptomeningeal disease, and the study presented by Smalley and colleagues represents an important next step in our understanding in how CSF can aid in both identifying the progression of disease as well as the development of resistance to targeted therapy in metastatic melanoma. Proteomic analysis, RNA sequencing, as well as both apoptosis and kinase assays showed significant differences between the patients with confirmed leptomeningeal disease and the control group. Intensifying the efforts to investigate this population and increasing the opportunities to obtain CSF in single or serial sampling, will be critical to advance our understanding of the complex biology and to develop more tailored therapeutic approaches for patients with leptomeningeal disease.

Disclosure of Potential Conflicts of Interest

I.C. Glitza Oliva is a paid consultant for and reports receiving other commercial research support from Bristol-Myers Squibb. H.A. Tawbi is a paid consultant for Bristol-Myers Squibb, Merck, Novartis, Genentech, and Array, and reports receiving commercial research grants from Bristol-Myers Squibb, Merck, Novartis, Genentech, Celgene, and GlaxoSmithKline. No other potential conflicts of interest were disclosed.

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References

1. Smalley I, Law V, Wyatt C, Evernden B, Fang B, Koomen JM, et al. Proteomic analysis of CSF from patients with leptomeningeal melanoma metastases identifies signatures associated with disease progression and therapeutic resistance. Clin Cancer Res 2020;26:2163–75.
2. Glitza IC, Smalley KSM, Brastianos PK, Davies MA, McCutcheon I, Liu JKC, et al. Leptomeningeal disease in melanoma patients: an update to treatment, challenges, and future directions. Pigment Cell Melanoma Res 2020 Jan 8 [Epub ahead of print].
3. Van Paemel R, Vlug R, De Preter K, Van Roy N, Speleman F, Willems L, et al. The pitfalls and promise of liquid biopsies for diagnosing and treating solid tumors in children: a review. Eur J Pediatr 2020;179:191–202.
4. Ma C, Yang X, Xing W, Yu H, Si T, Guo Z. Detection of circulating tumor DNA from non-small cell lung cancer brain metastasis in cerebrospinal fluid samples. Thorac Cancer 2020;11:588–93.
5. Boire A, Brastianos PK, Garzia L, Valiente M. Brain metastasis. Nat Rev Cancer 2020;20:4–11.

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