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Cell-free DNA next-generation sequencing liquid biopsy as a new revolutionary approach for arteriovenous malformation

Maria Palmieri, MS,a Aurora Currò, MD,a,b Andrea Tommasi, MD,a,b Laura Di Sarno, MS,b
Gabriella Doddato, MS,a Margheria Baldassarri, MD,b Elisa Frullanti, MS,a Ann Rita Giliberti, MS,a
Chiara Fallerini, MS,a Angelo Spinazzola, MD,c Anna Maria Pinto, MD, PhD,b Alessandra Renieri, MD, PhD,a,b
and Massimo Vaghi, MD,c,d Siena and Crema, Italy

ABSTRACT
Objective: Somatic mosaicism of KRAS gene is currently recognized as the only established molecular basis of arteriovenous malformations (AVM). However, given the limitations of the current technologies, KRAS somatic mutations are detected only in a limited proportion of AVMs and tissue biopsy remains an invasive high risky, sometimes life-threatening,diagnostic procedure. Next-generation sequencing liquid biopsy using cell-freeDNA(cfDNA) has emerged as an innovative
noninvasive approach for early detection and monitoring of cancer. This approach overcomes the space-time profile constraint of tissue biopsies opens a new scenario for vascular malformations owing to somatic mosaicism. Here, we propose a new approach as a fast noninvasive reliable tool in order to investigate the cfDNA coming from the AVMs.
Methods: A group of five patients suffering from AVM were selected. Blood samples from peripheral vein and efferent vein from vascular malformation were collected and cfDNA was extracted. The cfDNA libraries were performed using
Oncomine Pan-Cancer Cell-Free Assay. We used Ion Proton for sequencing and Ion Reporter Software for analysis (Life Technologies, Carlsbad, Calif).
Results: In all cases, either G12D or G12V mutations in KRAS were identified. The mutational load was higher in the efferent vein than in peripheral blood, confirming the causative role of the identified mutation at a somatic level.
Conclusions: We demonstrate that cfDNA next-generation sequencing liquid biopsy is able to identify the KRAS mutation detected in affected tissues. Moreover, we have shown that blood sample withdrawal at the lesion site increases variant allele frequency with an order of magnitude above the limit of detection (usually 0.05%), decreasing the risk of a
false negative. Finally, the noninvasiveness of the method avoids any risk of bleeding, being easily performed also in children. We propose this technique as the method of choice to better investigate AVMs and consequently to identify the therapy tailored to the genetic defect. (JVSeVascular Science 2020;1:176-80.)
Clinical Relevance: This article highlights the importance of using liquid biopsy as a new method to investigate the molecular profile of AVMs. In view of the frequent inaccessibility of vascular tissues owing to the invasiveness of solid
biopsy and the relative high incidence of biopsies with low diagnostic power, here we evaluated the efficacy of detecting cfDNA fragments released into the bloodstream from the affected tissue cells. Through a simple blood draw from the efferent vein at the vascular malformation site, the liquid biopsy allowed us to identify KRAS pathogenic mutations piloting a personalized therapeutic approach and opening a new scenario for new therapeutic strategies.
Keywords: Arteriovenous malformation; Liquid biopsy; cf-DNA; KRAS mutation; Noninvasive technique

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