What can a fluorescent Epstein-Barr virus tell us about host-shutoff?

Alejandro Casco[1], Makoto Ohashi[1], and Eric Johannsen[1,2]

[1] Department of Oncology, McArdle Laboratory for Cancer Research, University of Wisconsin, Madison, Wisconsin, USA; [2] Department of Medicine, Division of Infectious Diseases, University of Wisconsin, Madison, Wisconsin, USA

casco@wisc.edu

Epstein-Barr virus (EBV) reactivation from latency is characterized by increased lytic transcription, but also a dramatic loss of cellular mRNAs. This host shutoff is due to mRNA cleavage by the BGLF5 encoded nuclease as well as decreased transcription of cellular genes. While host shutoff is undoubtedly important for EBV's ability to overcome innate anti-viral responses, our knowledge of the specific mRNAs subject to shutoff is limited. A major limitation to addressing this gap in our knowledge is that only a few percent of latently infected cells can be induced to reactivate, and they proceed asynchronously through the lytic cycle. To address this, we have constructed an EBV reporter virus that expresses GFP with early kinetics and RFP with late kinetics. Coupling this system with fluorescence-activated cell sorting (FACS) enabled high-resolution transcriptomic profiling of lytic sub-populations and protein profiling at a single-cell level. We identified novel cellular changes that occur during the early and late stages of EBV replication, including host factors that may restrict or promote the completion of lytic replication. Our results have implications for how EBV reactivates without inciting an inflammatory response and may identify barriers to reactivation that can inform lytic induction strategies to treat EBV associated cancers.