Name: Dan Fabris

Email: dan.fabris@uconn.edu

Investigating epitranscriptomic communications between HIV-1 infection and neuromodulator response in different CNS cells

Limin Deng, Mikaila French, Jyotsna Kumar, and Dan Fabris

University of Connecticut, Department of Chemistry

RNA post-transcriptional modifications (rPTMs) have rapidly emerged as important factors mediating virus-host interactions during infection. At the same time, their regulatory roles in neurons and other CNS cells is progressively coming into focus. The extensive overlap between these epitranscriptomic functions suggests possible communications between RNA processes involved in viral replication and neuromodulator response. To identify such communications, we investigated the effects of neuromodulators, such as dopamine, GABA, glutamate, and histamine, on the identity and incidence of rPTMs expressed by primary human neurons, as well as primary astrocytes, microglia, and oligodendrocytes. Although the latter are not involved in signal transmission in the brain, they possess receptors for the selected neuromodulators and are capable of responding to their presence by exhibiting a rapid increase in cellular Ca2+. We capitalized on this characteristic to explore the expression of rPTMs as a function of time, which was assessed by mass spectrometric (MS) analysis. The results were integrated with those obtained from a series of assays aimed at monitoring fundamental cellular processes, in such a way as to enable their correlation with the observed rPTM expression patterns. The results revealed a subset of modifications that were significantly affected (in both positive and negative directions) by the neuromodulators, but reverted to normal levels within minutes to few hours. Others instead exhibited variations that persisted up to 48 h. The same experimental strategy was implemented also on an immortalized astrocyte cell line (i.e., human astrocytoma U251 MG), which was infected with HIV-1 and then treated with the selected neuromodulators at either 12 or 24 h post infection. The results led to the identification of rPTMs that were upregulated by the viral infection and further potentiated by neuromodulator treatment. We are now moving in two directions: a) confirm the observation in both primary human astrocytes and neurons; b) generate U251 MG knockdowns in which the biosynthetic enzymes for such rPTMs are effectively silenced. The results will place us closer to confirm the role of such rPTMs as communication hubs between HIV infection and neuromodulators acting on drugs of abuse pathways.