Components of calcium signaling in autophagy

Jennifer Giles MA[1], Abel Ferow BS[1], and Quang-Kim Tran, MD, PhD[1]
[1]Des Moines University, West Des Moines, IA

Macroautophagy (autophagy) is a process that packages and delivers unwanted cellular components to the lysosome for degradation to maintain homeostasis. Autophagy is strongly activated by nutrient starvation, such as found in myocardial ischemia or infarction. Starvation, mimicked experimentally by removal of extracellular amino acids, induces an increase in intracellular Ca^{2+} (operationally, SICS, *starvation-induced Ca*$^{2+}$ *signal*) that is predicted to initiate many Ca^{2+}-dependent activities of autophagy via interactions with its ubiquitous transducer calmodulin (CaM). The *transient receptor potential mucolipin* 1 (TRPML1) is an important lysosomal Ca^{2+} release channel that participates in organellar fusion and acidification, critical steps of autophagy. We recently showed that CaM is critical for autophagy. However, whether CaM regulates components of SICS and lysosomal Ca^{2+} release via TRPML1 is unknown.

Here, we report that SICS consists of both organellar Ca^{2+} release and extracellular Ca^{2+} entry. Molecular buffering of CaM using a fusion of two high-affinity CaM-binding proteins is associated with substantial increases in both components. Activation of TRPML1 using the agonist ML-SA1 triggers a small Ca^{2+} release signal and a large Ca^{2+} entry signals that are higher with CaM buffering at all ML-SA1 doses tested. Notably, increasing buffering of CaM in a multiplexed imaging system that simultaneously detects the CaM sequesters and measures Ca^{2+} responses is associated with increasingly larger lysosomal Ca^{2+} release via the TRPML1. These data clearly indicate that nutrient starvation triggers increases in intracellular Ca^{2+} via multiple sources and that CaM regulates all its components. The data also suggest that CaM may directly inhibit TRPML1.