Autophagy induction by histone deacetylase inhibitors inhibits HIV-1 infection in macrophages

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ABSTRACT

Background: Histone deacetylase inhibitors (HDACi) have been shown to induce HIV production from latently infected cells in vitro with the aim to enhance elimination of these cells and clear a cure. However, the activity of these drugs in mouse T-cell reservoirs is unknown. In this study, we investigated the effect of HDACi on the induction of autophagy, and its implication of HIV within human primary macrophages.

Methods: Monocyte-derived macrophages (MDM) were treated with HDACi (romidepsin, vorinostat, belinostat, givinostat, or panobinostat) at sub-lethal concentrations and assessed for autophagy flux using the expression and phosphorylation of autophagy proteins and the lipodification of microtubule-associated protein 1 light chain 3B (LC3B) by immunoblotting, ARF-6/7, and fluorescence microscopy. MDM were challenged with 105 TCID50 HIV-1Ba-L per 5 x 105 cells, and replication assessed using HIV p24 antigen release into culture supernatant. The role of autophagy in the HDACi-mediated inhibition of HIV infection was investigated using small molecule inhibitors or transducing MDM with shRNA for BECN1. Data were analyzed using the Mann-Whitney U test.

Results: HDACi induced autophagy in MDM in a dose-dependent manner (Belinostat < Vorinostat < Panobinostat < Romidepsin < Givinostat) as demonstrated by increased lipodification of LC3B, degradation of superoxide dismutase (SOD) and increased autophagosome formation (P < 0.05) in the absence of macrophage cell death. We also identified that HDACi induces autophagy through a ULK complex-dependent mechanism by suppressing MTOR activity as indicated by the dephosphorylation of RPS6KB2, EIF4EBP1 and ULK1, the most downstream autophagy pathway components. When MTOR is inhibited, ULK1 is assembled on endocytic membranes that intersect with recycling endosomes. The phosphorylation status of well known MTOR substrates ULK1, RPS6KB2 and EIF4EBP1, all upstream autophagy pathway components.

Conclusions: HDACi inhibit HIV-1 infection in macrophages by inducing autophagy.

BACKGROUND

HIV exploits autophagy for its replication in macrophages. HIV-1 is assembled on endocytic membranes that interact with recycling endosomes.

• Autophagy is a major self-degradative process in mammalian cells with roles in cellular homeostasis.
• The ULK1 complex is the most upstream component in the autophagy pathway and is essential for autophagy induction.
• MTOR suppresses autophagy by phosphorylating and inactivating the ULK1 complex. When MTOR is inhibited, ULK1 is dephosphorylated, released from inhibition and induces autophagy.
• Histone deacetylase (HDAC)-inhibitor therapy can be a new class of therapeutic agents with promising outcomes during the treatment of a wide range of cancer types.

RESULTS

Figure 1

HDACi inhibit HIV-1 infection in macrophages

Figure 2

HDACi induce autophagy in macrophages

Figure 3

HDACi inhibit the mammalian target of rapamycin (MTOR)

Figure 4

HDACi inhibition of HIV-1 requires autophagosome formation

Figure 5

HDACi inhibit HIV-1 through the induction of autophagic flux

CONCLUSIONS

• HDACi inhibit the replication of HIV-1 in macrophages.
• HDACi trigger autophagy by suppressing MTOR and activating the autophagy kinase ULK1.
• Belinostat, givinostat and vorinostat, and to a lesser extent, panobinostat and romidepsin inhibition of HIV-1 is dependent upon autophagy.

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