Background
Hepatitis B virus (HBV) capsid assembly is an attractive target for the treatment of chronic hepatitis B (CHB). Class I cap induction modulators (CAMs), also called CAM-A induce HBV core protein (HBc) aggregation and sustained HBc reduction in CHB mouse models.1, 2 The underlying mechanism governing this effect has not yet been elucidated. Here, we present an exploration of the mechanism of action of a reference CAM (CAM-A), RG7907.

Methods

RNA-sequencing of HBV-expressing HepG2.117 cells to RG7907 or compound B for up to 70 days, either in the presence or absence of doxycycline (no HBc expression) or in its absence (inducing HBc expression). Interestingly, unlike compound B, RG7907 induced a time- and dose-dependent CAM1-induced cell death (CCD), starting on day 21. This effect was observed only in the absence of doxycycline, confirming the dependence on both HC and RG7907. Annexin V and caspase 3/7 assays showed that RG7907-mediated CCD is likely mediated through apoptosis, in line with in vivo HCC and HBcHS data. Finally, we demonstrate that HBc is necessary and sufficient for CCD by using HBcHS NTCP cells expressing HA-Tagged HBc and L-HBsAg. Here, treatment of cells with RG7907 for only 7 days significantly induced apoptosis in HBc-expressing cells with no effect in HcHs-expressing cells.

Conclusions

Figure 1 - Top left: Western blot showing a dose-dependent loss of HBc expression in HepG2.117 cells incubated with RG7907 at 1 to 100 nM for 48 h. Figure 1 - Top right: Western blot showing a dose-dependent loss of HBsAg expression in HepG2.117 cells incubated with RG7907 at 1 to 100 nM for 48 h. Figure 1 - Bottom left: Western blot showing a dose-dependent loss of L-HBsAg expression in HepG2.117 cells incubated with RG7907 at 1 to 100 nM for 48 h. Figure 1 - Bottom right: Western blot showing a dose-dependent loss of NTCP expression in HepG2.117 cells incubated with RG7907 at 1 to 100 nM for 48 h.

Figure 2 - Figure 2 - Top left: Western blot showing a dose-dependent loss of HBc expression in HepG2.117 cells incubated with RG7907 at 1 to 100 nM for 48 h. Figure 2 - Top right: Western blot showing a dose-dependent loss of HBsAg expression in HepG2.117 cells incubated with RG7907 at 1 to 100 nM for 48 h. Figure 2 - Bottom left: Western blot showing a dose-dependent loss of L-HBsAg expression in HepG2.117 cells incubated with RG7907 at 1 to 100 nM for 48 h. Figure 2 - Bottom right: Western blot showing a dose-dependent loss of NTCP expression in HepG2.117 cells incubated with RG7907 at 1 to 100 nM for 48 h.

Figure 3 - Figure 3 - Top left: Western blot showing a dose-dependent loss of HBc expression in HepG2.117 cells incubated with RG7907 at 1 to 100 nM for 48 h. Figure 3 - Top right: Western blot showing a dose-dependent loss of HBsAg expression in HepG2.117 cells incubated with RG7907 at 1 to 100 nM for 48 h. Figure 3 - Bottom left: Western blot showing a dose-dependent loss of L-HBsAg expression in HepG2.117 cells incubated with RG7907 at 1 to 100 nM for 48 h. Figure 3 - Bottom right: Western blot showing a dose-dependent loss of NTCP expression in HepG2.117 cells incubated with RG7907 at 1 to 100 nM for 48 h.

Figure 4 - Figure 4 - Top left: Western blot showing a dose-dependent loss of HBc expression in HepG2.117 cells incubated with RG7907 at 1 to 100 nM for 48 h. Figure 4 - Top right: Western blot showing a dose-dependent loss of HBsAg expression in HepG2.117 cells incubated with RG7907 at 1 to 100 nM for 48 h. Figure 4 - Bottom left: Western blot showing a dose-dependent loss of L-HBsAg expression in HepG2.117 cells incubated with RG7907 at 1 to 100 nM for 48 h. Figure 4 - Bottom right: Western blot showing a dose-dependent loss of NTCP expression in HepG2.117 cells incubated with RG7907 at 1 to 100 nM for 48 h.

Figure 5 - Figure 5 - Top left: Western blot showing a dose-dependent loss of HBc expression in HepG2.117 cells incubated with RG7907 at 1 to 100 nM for 48 h. Figure 5 - Top right: Western blot showing a dose-dependent loss of HBsAg expression in HepG2.117 cells incubated with RG7907 at 1 to 100 nM for 48 h. Figure 5 - Bottom left: Western blot showing a dose-dependent loss of L-HBsAg expression in HepG2.117 cells incubated with RG7907 at 1 to 100 nM for 48 h. Figure 5 - Bottom right: Western blot showing a dose-dependent loss of NTCP expression in HepG2.117 cells incubated with RG7907 at 1 to 100 nM for 48 h.