Constructing an HBV Reporter Virus

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Hepatitis B virus (HBV), a small partially double-stranded DNA hepadnavirus, is the causative agent of B-type hepatitis. A replication-competent HBV vector bearing a reporter gene (reporter virus) would enable development of therapeutics to treat chronic HBV infection, but currently no such vector exists. HBV's small genome size and the need to preserve a number of overlapping reading frames is largely responsible for the difficulty in engineering such a vector. There are several strategies for generating a replication-competent HBV reporter virus that seeks to address these difficulties. Necessary genetic information present in overlapping reading frames was preserved by duplicating various regions of the HBV genome, which provided unique sites for reporter gene insertion. Anticipating that the sites of insertion may be suboptimal for HBV replication, a method was devised to exploit the high mutation rate of HBV polymerase and harness the power of natural selection. To this end, the initial insertions encode for a blasticidin resistance gene (Bsd), thereby providing cells that replicate the HBV reporter with resistance to blasticidin. Ongoing work aims to identify adaptive mutations and reintroduce them into the HBV reporter vectors. From here, both fluorescent and luminescent reporter genes will be inserted and the resulting HBV vectors will be characterized.