Hepatitis B virus replication in primary macaque hepatocytes: crossing the species barrier toward a new small primate model.

The development of new anti-hepatitis B virus (HBV) therapies, especially immunotherapeutic approaches, has been limited by the lack of a primate model more accessible than chimpanzees. We have previously demonstrated that sylvanus and cynomolgus macaques are susceptible to in vivo HBV infection after intrahepatic HBV DNA inoculation. In this study, we evaluated the susceptibility of primary macaque hepatocytes (PMHs) to HBV infection with a highly efficient HBV genome-mediated transfer system via a recombinant baculovirus (Bac-HBV). Freshly prepared PMHs, isolated from macaque liver tissue by collagenase perfusion, were transduced with Bac-HBV, and intermediates of replication were followed for 9 days post-transduction. Evidence of HBV replication (hepatitis B surface antigen secretion, viral DNA, RNA, and covalently closed circular DNA) was detected from day 1 to day 9 post-transduction. HBV markers were dose-dependent and still detectable at a multiplicity of infection of 10. Importantly, transduced PMHs secreted all typical forms of HBV particles, as evidenced by a cesium chloride gradient as well as transmission electron microscopy. Furthermore, the Toll-like receptor 9 (TLR9) ligand was used to stimulate freshly prepared macaque peripheral blood mononuclear cells to generate...
TLR9-induced cytokines. We then demonstrated the antiviral effects of both TLR9-induced cytokines and nucleoside analogue (lamivudine) on HBV replication in transduced PMHs. CONCLUSION: Baculovirus-mediated genome transfer initiated a full HBV replication cycle in PMHs; thus highlighted both the baculovirus efficiency in crossing the species barrier and macaque susceptibility to HBV infection. Moreover, our results demonstrate the relevance of this system for antiviral compound evaluations with either nucleoside analogues or inhibitory cytokines. Cynomolgus macaques are readily available, are immunologically closely related to humans, and may therefore represent a promising model for the development of new immunotherapeutic strategies.