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Inhibition of Duck Hepatitis B Viral Replication by Conventional Inhibitors and Supercoiled DNA Active Compounds. G Civitico, Y Wang, G Tachedjian, I Gust and S Locarnini. Macfarlane Burnet Centre for Medical Research, Fairfield Hospital, Fairfield 3078, Victoria, Australia.

One of the key events in hepadnaviral replication is the conversion of relaxed circular virion DNA to the supercoiled (SC) form in the host cell nucleus, where it then acts as the functional template for transcription. This replicative form of the viral DNA has proven to be resistant to current antiviral therapies so that any SC DNA that is not eliminated by treatment can act as a reservoir for re-initiation of replication. To understand this process in more detail we asked the question: How is hepatitis B virus (HBV) SC DNA generated and processed? As part of this investigation we tested a range of compounds for their effects of duck hepatitis B viral (DHBV) replication in congenitally infected primary duck hepatocyte (PDH) cultures. DHBV is morphologically and genetically similar to HBV. PDH were treated with a range of compounds including conventional agents, reverse transcriptase inhibitors and compounds with specific activities directed to SC DNA [topoisomerase and DNA gyrase inhibitors] and then analysed for viral DNA replicative intermediates and protein production. This study identified 8 compounds which significantly inhibited viral replication: the conventional inhibitors ganciclovir, acyclovir, ribavirin and phosphonoformate, the topoisomerase II inhibitors amsacrine and ellipticine and the DNA gyrase inhibitors coumermycin A1 and nalidixic acid. The conventional inhibitors had no effect on DHBV SC DNA and viral protein production was also unaffected. The SC DNA active compounds reduced the amount of viral SC DNA present in cells and significantly inhibited viral DNA and protein generation. In conclusion, we have identified a new class of compounds which can inhibit DHBV replication in chronically infected PDH which could have significance for the management of human hepatitis B infection.

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Synthesis and Antiviral (RNA) Evaluation of Nucleoside Analogs of Ribavirin and Tiazofurin Modified at the Carboxamide Moiety.

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A series of ribavirin and tiazofurin analogues was synthesized wherein the carboxamide moiety was replaced by the following functionalities: a) mono- and di-N-alkylated carboxamidines; b) carboxamidines coupled to the amino acids glycine, asparagine and glutamine; c) N-cyanocarboxamidine; d) methyl and ethylcarboximidates; and e) 1,4,5,6-tetrahydropyrimidines. These compounds were evaluated <u>in vitro</u> against the RNA-containing flaviviruses (Japanese encephalitis, yellow fever and dengue fever viruses), bunyaviruses (Punta Toro, sandfly fever and Rift Valley fever viruses) and the alphavirus (Venezuelan equine encephalitis virus). Several were also evaluated against HIV-1. Antiviral activity was observed with the mono- and di-N-alkylated carboxamidines and the glycine-coupled carboxamidine of ribavirin. This activity was predominantly limited to the bunyaviruses and, in some cases, to dengue fever virus.