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A Variant of Echovirus 12 that Requires the Selective Antiviral Inhibitor Rhodanine for Replication. H.J. Eggers, J.P. Kruppenbacher, Institut fuer Virologie, Universitaet zu Koeln, D-50935 Koeln, Germany

Rhodanine, 2-thio-4-ketothiazolidine, is the first selective antipicornaviral inhibitor with capsid-binding properties studied in detail (Eggers, H. J. (1977) Virology **78**: 242-252). It inhibits exclusively the uncoating step in the replication cycle of echovirus 12 by stabilization of the virion. Very early we have isolated rhodanine-resistant and -dependent mutants. The properties of one dependent echovirus 12 mutant will be reported. In this case, rhodanine is needed only for attachment to cells at room temperature or at 37°C. All further steps such as uncoating and virus synthesis do not require the compound. Attachment at 4°C is also drug-independent, but warming up to room temperature leads to a rapid loss of potentially infectious virus from host cells. The drug-dependent mutant appears at least as thermostable as the wild type, and up to 44°C there are no significant differences in inactivation kinetics regardless whether rhodanine is present or not. We shall compare the properties of our mutant to virus variants dependent on other capsid-binding inhibitors. The experiments appear useful to elucidate processes in early virus-cell interactions.

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Inhibition of Hepatitis B Virus Replication in vitro by Antisense Oligonucleotides. B. Korba, F. Wells, K. Jones, R. Engle, A. Buckler-White, and J. Gerin. DMVI, Georgetown Univ. Medical Center, Rockville, MD USA

The majority of antiviral approaches against Hepatitis B Virus (HBV) are focused on the use of nucleoside analogues to inhibit viral polymerase activities. Antisense oligonucleotides are currently being utilized by numerous laboratories as anticancer and antiviral agents. The unique replication cycle of HBV is especially amenable to modulation by antisense oligonucleotides, which could potentially interfere with several different steps in the viral replication pathway. We have examined the ability of 50 different single-stranded, deoxyribonucleotides (14-23 nucleotides long), which target several HBV-specific functions, to inhibit HBV replication in the human hepatoblastoma cell line, 2.2.15. Oligonucleotides directed against the HBV surface antigen (HBsAg) gene (S gene) and the preS1 coding region inhibited HBsAg and virion release by the cells, but did not affect intracellular HBV replication. Antisense molecules which were targeted to within 20-25 bases of the AUG of the S gene were the most effective at inhibiting HBsAg production. Oligonucleotides directed against the HBV core antigen (HBcAg) gene depressed virion production and intracellular levels of HBcAg and HBV DNA replication intermediates. Oligonucleotides directed against the coding region for HBV e antigen (HBeAg) and the HBV polymerase gene did not affect the levels of HBV replication, although molecules directed against the HBeAg coding region depressed the levels of HBeAg released by the cells. Oligonucleotides directed against the upper stem-and loop of the HBV encapsidation signal/structure, were the most effective inhibitors of HBV replication. None of the oligonucleotides examined affected the levels or the sizes of the associated HBV-specific RNA transcripts indicating that the primary mechanism of action of these molecules was through an inhibition of RNA translation. A lack of antiviral activity by a series of "sense" oligonucleotides to various HBV genes indicated that the oligonucleotides were acting in a sequence-specific manner. Antisense oligonucleotides represent a promising approach to anti-HBV therapy which can be used to modulate several different steps in the HBV replication cycle.