Oral Session V: Hepatitis Viruses II, Herpesviruses II and Poxviruses II

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The Cyclophilin Inhibitor Debio-025 is a Potent Inhibitor of Hepatitis C Virus Replication in vitro With a Unique Resistance Profile

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Debio-025 is a potent inhibitor of HCV replication [Hepatology 43:761–770]. In phase I, clinical studies monotherapy (dose 1200 mg BID) resulted in an average viral load reduction of 3.6 $\log_{10}$ [Hepatology, 44: 4S1, 609A). We now demonstrate that Debio-025 is equipotent against wild-type HCV, as against HCV replicons that are resistant to either 2′-C-methylcytidine or the protease inhibitors VX-950 or BILN-2061. Debio-025, alone at concentrations below 1 μM, was able to cure cells from their HCV replicon within three to four passages, whereas treatment with a HCV protease inhibitor, (seven passages) did not. Debio-025, at a concentrations of 0.1 μM when combined with VX-950 [at a concentration that is alone not able to clear replicon cells] was able to efficiently cure the cells. Debio-025 at 0.1 or 0.5 μM was able to completely prevent the development of BILN-2061 and VX-950 resistant replicons. Following long-term culture (respectively, 28 and 52 passages) in increasing concentrations of Debio-025 or CsA, replicon resistance to both compounds was obtained. CsA and Debio-025 proved cross-resistant, but the replicons remained fully susceptible to interferon and various HCV polymerase and protease inhibitors. Stable transfection of naive Huh Lunet cells with CsA^r or Debio-025^r replicon RNA, resulted in cells that carry the drug-resistant phenotype, indicating that drug-resistance is replicon- and not host cell associated. Resistant replicons carry mutations restricted to one particular NS-gene [one AA-substitution for the CsA^r mutant and the same AA substitution plus a second one for the Debio-025^r mutant]. Because of its unique mechanism of action and resistance profile, Debio-025 may form an attractive drug candidate for the treatment of HCV infections in combination with polymerase and/or protease inhibitors.

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The Imidazopyrrolopyridine Analogue AG110 is a Novel, Highly Selective Inhibitor of Pestivirus Replication and Targets the viral RNA-dependent RNA Polymerase

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We report on the potent and selective anti-pestivirus activity of ethyl 2-methylimidazo[1,2-*a*]pyrrolo[2,3-*c*]pyridin-8-carboxylate (AG110). The EC_{50} for inhibition of bovine viral diarrhea virus (BVDV): (i) induced CPE formation is 1.2 ± 0.5 μM, (ii) RNA synthesis is 5 ± 1 μM and (iii) production of infectious virus is 2.3 ± 0.3 μM. AG110 is inactive against the hepatitis C virus and a flavivirus. Time-of-(drug)-addition experiments revealed that AG110 inhibits BVDV replication at a time point that coincides with the onset of intracellular viral RNA synthesis. Drug-resistant mutants ($AG110^r$) carry the E291G mutation in the viral RNA-dependent RNA polymerase (RdRp). $AG110^r$ virus is cross-resistance with the cyclic urea compound 1453 (Sun et al., 2003. J. Virol. 77, 6753–6760) which also induces the E291G mutation. Interestingly, $AG110^r$ virus is markedly less susceptible to inhibition by the imidazopyridine BPIP (Paeshuyse et al., 2006. J. Virol. 80, 149–160) a compound that induces the F224S resistance mutation. AG110 is a weak inhibitor of the in vitro activity of recombinant BVDV RdRp but inhibited efficiently the activity of BVDV replication complexes. Molecular modelling revealed that E291 is located in a small cavity near the tip of the finger domain of the RdRp about 7 Å away from F224. Docking of AG110 in the crystal structure of the BVDV RdRp revealed several potential contacts including with Y257. The E291G mutation may enable the free rotation of Y257; thus destabilizing the backbone of the loop 223–226 rendering more mobility to F224 and hence reducing the affinity for BPIP. The fact that also the pestivirus inhibitor VP32947 (Baginski et al., 2000. PNAS 97, 7981–7986) targets the viral polymerase and induces the F224S mutation, suggests that the larger E291/F224 region may be critical in the functioning of the polymerase or in the assembly of pestivirus replication complex.

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