Oral Presentations

New drugs for acute and chronic Hepatitis B Virus (HBV) Infection: from HBV-entry inhibition to liver-specific drug targetting

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The currently approved treatment options for chronic hepatitis B either obstruct HBV genome replication (nucleoside or non-nucleoside reverse transcriptase (RT) inhibitors) or stimulate innate or adaptive immune responses (interferon α (IFNα)). However, although these drugs are capable of reducing serum titres by several logs, elimination of the virus is commonly not achieved. Furthermore, prolonged virus replication at reduced levels in the presence of RT-inhibitors results in the selection and amplification of resistant mutants. This relates to the reservoir function and the high stability of cccDNA, the template of HBV transcription which accumulates in the nucleus of infected hepatocytes4. Since reduction of virus titres lowers the risk of developing liver damage and hepatocellular carcinoma, prolonged treatment even without achieving sustained virological responses is recommended.

In addition to the recent successes in the development of potent nucleoside analogues showing better resistance profiles or INFs with improved pharmacokinetic properties and less side effects, there are several promising new compounds that target so far unaddressed replication steps of HBV. One of these steps is nucleocapsid assembly which can be efficiently inhibited by a family of small molecules called dihydroarylpyrimidines (HAPs)1. HAPs, at low concentrations bind yet unassembled HBV core proteins thereby preventing the formation of an RNA-containing nucleocapsid and redirect the complex into a proteasome-dependent degradation pathway. Another approach addresses the stability of HBV-specific transcripts. Adenoviral transfer of HBV-specific of shRNAs into hepatocytes of HBV transgenic mice lead to a profound and sustained reduction of HBV gene expression3.

We have recently shown that large envelope protein-derived lipopetides efficiently interfere with HBV entry into hepatocytes in vitro and in vivo2. The lead substance of these peptides (Myrcludex B) consists of the first 47 amino acids of the HBV L-protein and is N-terminally myristoylated. Its IC50 in vitro is ~200 pM. Pharmacokinetic studies showed that the peptide targets the liver of even non HBV-susceptible animals with extraordinary selectivity, suggesting the presence of a species-independent but hepatocyte-specific receptor. Mutational analyses revealed that both, myristoylation and a conserved seven amino acid sequence motif are necessary for liver-targeting. Preclinical toxicology studies of Myrcludex B are ongoing and will be presented. Considering the possibility that even under strong suppression of viral replication infection of naive hepatocytes takes place, it will be interesting to see whether an entry inhibitor is capable of reducing the viral cccDNA in chronically infected patients.

Beside their direct antiviral activity the strong liver-tropism of non-immunogenic HBV surface protein derived lipopetides variants can also target other drugs to the liver. One approach is the delivery of IFNα to avoid systemic side effects. First results on the feasibility of this concept will be presented.

Taken together there are promising new substances under preclinical development. In the near future these drugs will hopefully broaden the therapeutic spectrum to treat chronic hepatitis B with increased success rates.

Next Generation Sequencing offers new perspectives in Comparative Genomics of plant associated Bacteria

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Next Generation Sequencing technologies have started to change genome research. Highly automated systems provided by companies like 454 Life Sciences or Illumina introduced a revolution in genome research. The power of the new technologies can be characterized by a few catchwords like: comparable low costs, high amount of data and short turn around time.

However, just a draft sequence can be provided by this approach for the majority of genome projects with de novo character in contrast to projects with re-sequencing character. Uncertainties resulting from the used technique and/or resulting draft sequences limit the benefit. In consequence, sequence quality has to be increased by the combination of new and old technologies. Examples and preliminary results from running projects will be presented to illustrate the use and benefit of the new techniques in the genome research of plant associated species from the Enterobacteriaceae and Acholeplasmataceae.

Oral Session I

Elucidation of the roles of blackcurrant reversion virus and phytoplasma in the etiology of full blossom disease in currants

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To determine the roles of phytoplasmas and Blackcurrant reversion virus (BRV) in the etiology of full blossom disease (FBD), we conducted graft and dodder transmission experiments. Scions from FBD affected Ribes rubrum were grafted onto red, white and black currants. Red and white cultivars revealed symptoms of FBD, whereas blackcurrant displayed symptoms of BRV infection. No differences in symptoms were observed between plants infected with BRV only and those infected with BRV and phytoplasma.

Aster yellows phytoplasma subgroup 16SrI-C was transferred from FBDinfected red currants to periwinkle, which symptoms of green and yellow petal were observed. Back transmission of phytoplasma to currant seedlings of red and black currant was not successful.

Scions of periwinkle infected with aster yellows phytoplasmas of subgroup 16SrI-C and 16SrI-B, which were bottle-, bark-, and approach-grafted onto seedlings of red and black currant resulted in positive but symptomless transmission of phytoplasma to red currant. We conclude that FBD symptoms are induced by BRV rather than by phytoplasma, which was originally described as the causal agent of FBD.

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Association of Tomato Rinsgpot Virus, Tobacco Ringspot Virus and *Xiphinema americanum* with a decline of highbush blueberry in New York

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A survey of highbush blueberry (Vaccinium corymbosum L.), including some cultivars showing virus-like symptoms, i.e. stunted growth, chlorotic spots, purple lesions, distorted leaves and defoliated shoots, was conducted for the occurrence of viruses in New York. Leaves from symptomatic and nonsymptomatic plants were tested for virus infection by enzyme-linked immunosorbent assay using specific serological reagents (Bioreba, Reinach, Switzerland). Several samples reacted positively for Tobacco ringspot virus (TRSV) and Tomato ringspot virus (ToRSV), two virus species belonging to the genus Nepovirus in the family Comoviridae. The occurrence of TRSV and ToRSV was confirmed in blueberry leaf samples by reverse transcription-polymerase chain reaction (RT-PCR) or immunocapture-RT-PCR with appropriate primers to amplify a 310-bp and a 580-bp fragment of the RNA-dependent RNA polymerase gene, respectively. Comparative sequence analysis of the viral amplicons of New York isolates indicated moderate to high nucleotide sequence identities with corresponding ToRSV and TRSV reference strains. Also, analysis of soil samples collected from the root zone of blueberry bushes for the occurrence of nematodes indicated the presence of specimens from the Xiphinema americanum group.