correspondently. Modulating influences of the lipotropic (cage-hydrocarbon) pharmacophores on the anti-CMV activity were observed only under the viricidal and prophylactic experimental schemes, where the lipid membranes of cells and/or viral envelope are involved. But the dominant role in the AVA antiviral activity was played by the sulf-anionic modulation. The negative charge accumulation on AVA macromolecules, seems, amplifies a potential for electrostatic blocking virions/virus-cell adsorption, and agonistic stimulated cell resistance or antagonistic competition of this synthetic polyacids with the viral nucleic acids. The most promising compounds have been selected for the future explorations of mechanisms of the anti-CMV and anti-HIV activity.

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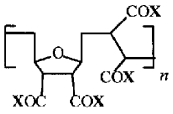

Where -X =
-OH/-O⁻ Na+, carboxyacid (CA), in part negative charged
-NH-Spacer$_1$-Adamantane (Ad), cage-tricyclic, mebranotropic
-NH-Spacer$_2$-exo-Norbornene (Nb), cage-bicyclic, mebranotropic
-O-Spacer$_3$-SO$_3^-$ Na+, sulfoacid (SA), full negative charged

AVA code	Various kind side groups (X), mol. ratio, CA : Ad : Nb : SA	Cytotoxicity [a], CC_{50}, μg/ml		Selectivity Index of Anti-CMV[b] activity SI = CC_{50}/EC_{50} (3 days)		
		1 day	3 days	Viricid.[c]	Prevent.[d]	Therap.[e]
ÀS. 470	1.00 : 0.00 : 0.00 : 0.00	3 750	3 500	< 10	< 10	< 10
ÀS. 473	0.94 : 0.06 : 0.00 : 0.00	3 200	2 500	< 10	< 10	< 10
ÀS. 632	0.93 : 0.07 : 0.00 : 0.00	3 600	2 400	41	< 10	< 10
ÀS. 504	0.92 : 0.00 : 0.08 : 0.00	3 200	1 700	< 10	< 10	< 10
ÀS. 677	0.86 : 0.00 : 0.08 : 0.06	-	1 440	66	22	< 10
ÀS. 678	0.79 : 0.00 : 0.08 : 0.13	1 800	1 420	355	189	< 10
ÀS. 679	0.67 : 0.00 : 0.08 : 0.25	1 000	500	**5 000**	91	< 10
ÀS. 688	0.60 : 0.00 : 0.00 : 0.40	4 000	3 000	**7 500**	**250**	**4 286**

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A Mass Spectrometry-based Method to Detect Antiviral Drug Resistance in Human Cytomegalovirus

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During antiviral therapy, the emergence of viral escape mutants that are resistant against the drug of choice is a major problem. Monitoring the introduction of resistance-associated mutations into the viral genome population can facilitate antiviral therapy management, including the selection of the most effective drug(s) in a given situation. We are currently assessing whether a mass spectrometry (MS)-based re-sequencing method (iSEQ by Sequenom, Inc., San Diego, USA) can improve the accuracy, speed, and sensitivity of the identification of resistance-associated mutations in the human cytomegalovirus genome (HCMV). Briefly, the assay employs four base-specific cleavage reactions of an amplicon of a relatively small region of the viral genome. The resulting four MS data sets are compared to *in silico* derived cleavage patterns from a database of reference sequences. Differences between the spectra derived from patient samples and those derived from viral reference sequences are indicators of sequence variations and can reveal potential resistance mutations. As resistance mutations against ganciclovir frequently occur in the phosphotransferase gene of HCMV (UL97), this gene was chosen to obtain proof of principle. A collection of patient samples was used to produce amplicons of 300–700 base pairs representing the 3′ half of the gene, which were analyzed using the Sequenom approach. The results were verified by traditional sequence analysis of the same samples. Our first data confirmed that detection of mutations or polymorphisms by SNP discovery is faster, but equally accurate compared to identification by regular sequencing. A more extensive comparison will be performed in the coming months.

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L-Analogs of 1-Beta-D-Ribofuranosyl-2-Bromo-5,6-Dichlorobenzimidazole (BDCRB) Inhibit Human Herpesvirus-6 Replication

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Benzimidazole nucleoside analogs have proven to be an abundant source of molecules with highly specific antiviral activity against human cytomegalovirus (HCMV). One analog, 1H-β-D-ribofuranosyl-2-bromo-5,6-dichlorobenzimidazole (BDCRB) inhibits the packaging of viral DNA, whereas a related L-ribosylnucleoside, 1H-β-l-ribofuranosyl-2-isopropylamino-5,6-dichlorobenzimidazole (maribavir) inhibits the HCMV UL97 protein kinase and is currently in Phase III clinical trials. Human herpesvirus-6 (HHV-6) is a related betaherpesvirus that is inhibited to a limited extent by maribavir but is insensitive to BDCRB. Therefore we hypothesized that other L-sugars in this series would be specific inhibitors of HHV-6. Of several compounds tested, two L-analogs of BDCRB (L-ribosyl BDCRB and (−)-carbocyclic BDCRB) have been identified that have good activity against the A variant of HHV-6 (EC_{50} = 2.6 and 2.4 μM, with selective indices of 11 and 5, respectively). Both compounds also inhibited HCMV in this concentration range (EC_{50} = 1.3–3.8 μM). These data differ with results for D-ribosyl analogs that were active against HCMV, but not HHV-6. Additional studies were conducted to examine their mechanism of action. Neither compounds inhibited viral DNA synthesis at high multiplicities of infection and no inhibition of HHV-6 U69 kinase activity was detected. Both results are consistent with a mechanism of action that is similar to that of BDCRB against HCMV and suggest that certain L-benzimidazole analogs have a mechanism of action similar to the D-benzimidazole analog BDCRB and differ from that of the L-analog maribavir. The results substantiate the prior observation that both the sugar moiety and the substituent in the 2-position of the heterocycle affect the mechanism of action and antiviral specificity of benzimidazole nucleosides.

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Comparative Efficacy of Treatment with CMX001 Versus Acyclovir in BALB/c Mice Infected with Herpes Simplex Virus

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Previous reports have shown excellent activity of CMX001 both in vitro and in vivo against vaccinia virus, cowpox virus, cytomegalovirus (CMV) and herpes simplex virus, Type 1 and 2 (HSV-1 and HSV-2). In cell culture, CMX001 has proven to be