172

QSAR Analysis of Anti-influenza (A/H1N1) Activity of Azolo-adamantanes

Eugene Muratov[1,2,*], Ekaterina Varlamova[1], Anatoly Artemenko[1], Victor Kuz'min[1], Pavel Anfimov[3], Vladimir Zarubaev[3], Victor Saraev[4], Oleg Kiselev[3]

[1] *A.V. Bogatsky Physical-Chemical Institute NAS of Ukraine, Odessa, Ukrenia*
[2] *University of North Carolina, Chapel Hill, USA*
[3] *Influenza Research Institute, Saint Petersburg, Russia*
[4] *Saint Petersburg State Institute of Technology, Saint Petersburg, Russia*

Keywords: Azolo-adamantanes; Influenza A/H1N1; Selectivity index

Influenza spreads around the world in seasonal epidemics, resulting in the deaths of up to 500 thousand people every year. Since vaccination and existing antivirals cannot guarantee protection against influenza, battling this virus remains important health care task that requires design and development of new drugs. Application of cheminformatics methods shortens the development time and reduces costs of antiviral drug research. The goal of the present study is computer-assisted design of novel selective agents by the means of QSAR analysis of antiviral activity of azolo-adamantanes against influenza. The dataset comprised 60 azolo-adamantanes. Thorough investigation of the relationship between antiviral activity against influenza strain A/PR/8/34 (H1N1) (EC_{50}, μM), cytotoxicity on MDCK cells (CTD_{50}, μM), and selectivity index (ratio of CTD_{50} to EC_{50}) and the structure of investigated compounds was carried out using Hierarchic QSAR Technology (HiT QSAR). Prior to development of QSAR analysis, the compounds were divided on two classes according to their activity, selectivity, and cytotoxicity: EC_{50}active < 0.1 μM < EC_{50}inactive CTD_{50}toxic < 1 μM < CTD_{50} non-toxic and SI non-selective < 10 < SI selective. Five-fold external cross-validation was used for the estimation of predictive power of obtained random forest models. We succeeded to develop predictive model of antiviral activity with cumulative classification correct rate CCR_{5FECV} = 0.8. The quality of cytotoxicity and selectivity models was somewhat lower but still acceptable (CCR_{5FECV} = 0.64–0.7). New selective anti-influenza agents were computationally designed and predicted using developed models. Six of them were recommended to synthetic and biological experiments.

doi:10.1016/j.antiviral.2011.03.158

173

Viral Genome Dynamics During Antiviral Resistance Selection: A First Glimpse into Viral Evolution

Simone Musiu*, Jan Paeshuyse, Mathy Froeyen, Philippe Lemey, Johan Neyts

Katholieke Universiteit Leuven, Leuven, Belgium

The nature of the RNA-dependent RNA polymerase (RdRp) of most RNA viruses renders their genome prone to the accumulation of mutations. Hence the virus population exists as a complex and dynamic mutant distribution, i.e. a quasispecies. The quasispecies enable rapid adaptation of the virus to any changes in its natural environment. Also during antiviral therapy, rapid selection of drug resistant virus is facilitated by the existence of the virus as a quasispecies rather than a defined genomic sequence. We employed the bovine viral diarrhea virus (BVDV) [family of the *Flaviviridae*, genus pestivirus] as a model virus. Most, if not all of the currently know pestiviral RdRp inhibitors target a 7 Å region between F224 and E291 within the finger domain of the enzyme. Here we describe how the RdRp coding region of the viral genome evolves during *in vitro* antiviral resistance selection. To this end we employed a panel of selective inhibitors of BVDV replication i.e. LZ37 (Paeshuyse et al., 2009), AG110 [Paeshuyse et al., 2007) and BPIP (Paeshuyse et al., 2006). Resistant virus was selected (against each compound) by serially passaging (25 times) the virus in the presence of increasing concentrations of inhibitor. The entire NS5B gene was sequenced. Furthermore the sequence flexibility of the polymerase region F224–E291 was analysed. It was observed that different resistance mutations could be obtained during independent parallel resistance selection. The sequence of BPIP- and LZ-resistant BVDV clustered separately from AG110-resistant BVDV. This might indicate that there exist certain genomic constraints that impede the potential truly random nature of the quasispecies. To study this we designed a resistance selection scheme for AG110 and BPIP that allowed monitoring genomic changes in parallel. Preliminary data highlight a more complex evolutionary pattern than initially observed. The results provide a first glimpse into patterns of viral evolution during selective antiviral pressure exerted by specific pestiviral polymerase inhibitors.

References

Paeshuyse, et al., 2009. Antivir. Res. 82, 141–147.
Paeshuyse, et al., 2007. J. Virol. 81, 11046–11053.
Paeshuyse, et al., 2006. J. Virol. 80, 1469–1600.

doi:10.1016/j.antiviral.2011.03.159

174

Structure-based design of small-molecules that selectively inhibit dengue virus methyltransferase

Christian G. Noble*

Novartis Institute for Tropical Diseases, 10 Biopolis Road, Singapore

Flaviviruses contain a single methyltransferase (MTase) for methylation of the 5′ cap of the Flavivirus RNA. This protein contains a single domain with both guanine N7 and ribose 2′O MTase activities. We have analysed the crystal structures of Flavivirus MTases and identified a Flavivirus-conserved hydrophobic pocket next to the binding site for S-adenosyl-methionine (SAM), the substrate of the methylation reaction. Several derivatives of S-adenosylhomocysteine (SAH), the product of the methylation reaction, were synthesised. These compounds were designed to contain substituents that extended into the hydrophobic pocket and were found to be more potent against dengue virus MTase than SAH, but were not active against related human enzymes. Crystal structures showed that these compounds bound into the hydrophobic pocket of dengue MTase, and induced conformational changes in residues lining the pocket. The structures show the specific interactions between the compounds and the dengue MTase and suggest ways to further improve their potency. Together these data show that selectivity for disease-related MTases, can be produced.

doi:10.1016/j.antiviral.2011.03.160