56
Inverse Wavelet Transform in Virus–Cell Interaction Imaging

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We have shown previously that the diffraction patterns obtained by laser irradiation scattering on virus–cell system demonstrate fractal properties which reflect the fractal structure of the system itself. Diffraction process is equivalent to the formation of the wavelet direct Fourier transform of the said system's components, including sensitive cells (about 10 μm n diameter), cell nuclei (about 1 μm) and virions (about 0.1 μm). The multi-order imaging of the virus–cell system is provided due to the self-affinity of the fractal aggregates involved in imaging process. We propose here to use the inverse wavelet Fourier transform of the pattern formed on the target of the fractal microscope in order to get the real enlarged image of the viruses attacking the sensitive cell as well as the cell's structural transformation caused by the interaction itself. The set of bright and dark spots, which forms the diffraction pattern, could be transferred into set of numbers using the regular quantification procedure. The full information included into the pattern peaks' diameters and color index is transformed using inverse wavelet Fourier technique into set of intersecting bright and dark circles. The full *in vitro* dynamics of the structural changes of the virus–cell system is described by the changes of circles' diameters and their intersection's area. It was shown, also, that the magnification of the proposed fractal microscope could achieve 10,000–100,000×, depending on the used laser power. Proposed fractal microscope could be applied as well *in vivo* experiments until the required magnification will not make us to use projection laser with the output exceeding 20 mW. The reliability and sensitivity of the proposed device is defined by the parallel virus–cell structural information processing by a regular laptop. The fractal microscope based on the inverse wavelet Fourier transform procedure could be applied successfully in pharmaceutical antiviral drug design, laboratory and clinical trials of new antiviral preparations.

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57
New Microscopic Description of Herpes Virus–Cell Dynamic System

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We have shown that the fractal approach to the problem of virus–cell interaction gives the unique possibility to process the data through the sequence of the direct and inverse wavelet Fourier transforms. We have studied the Herpes simplex virus US-1 strain interacting with the Hep-2 sensitive cell culture. The object was imaged as bright peaks considered as wavelets formed as a result of a laser diffraction on the structural elements of the virus–cell system. The whole virus–cell interaction information is inserted into computer in a fastest parallel way. The laser intensity peaks, forming the speckle image of the system under consideration, could be transformed into the hierarchical system of the circles (or squares) according to the choice of the researcher, but conserving the same D value, which depends only on the true intermolecular interaction potential. This potential, being characteristic for every stage of virus–cell interaction, is responsible also for the structure of the dynamic virus–cell system. The unique, but the typical form of the fractal cluster corresponding both to the system itself and its image as well, was processed by computer techniques. The hierarchical fractal design of the virus–cell system, proposed here for the first time, gives the universality, needed for the quantitative description of any possible combination of the virus and corresponding sensitive cell. It should be noted, as well, that the fractal microscope use for virus–cell dynamic system imaging have all the properties, required from all other experimental tools of monitoring, including the reliability, reproducibility and preciseness. The device could be used in drug design tests with the scope of time and efforts economy during the compounds libraries screening. The fractal microscope combined with the QSAR drug design technique makes the anti-herpetic drug design more competitive as compared to the regular approaches.

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58
Dielectric Spectroscopy as a Tool for Virus–Cell Interaction Rate Description

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We propose to apply the dielectric spectroscopy (DS) of virus–cell interaction in laboratory practice. We have chosen the human immune deficiency virus (HIV) applied to the lymphocytes system as the actual object for investigation at various stages of interaction. The thin layer (10 μm) of the sensitive cells (SC) placed between parallel glass plates with transparent SnO_2–In_2O_3 electrodes. HIV was added in concentration of 10^{12} m^{-3} what means about 1 ppm ratio. The DS curves for real ε' and imaginary ε'' parts of the complex dielectric permeability were registered for the chamber containing either SC only or with addition of HIV particles with the use of BM-560 Q-meter in the frequency range of 50 kHz–35 MHz. We have shown experimentally that the DS curves demonstrated the presence of various number of maxima depending on the

type of the substrate as well as on the presence of HIV particles and their concentration. We have seen the changes of maxima heights and their half-widths. The DS curves have shown in experiment the non-Debye behavior what could be attributed to the fractal clustering process occurring and changing at various stages of virus–cell interaction. The maxima registered for SC sample were sheared to the lower frequency region after the time needed for viral nucleic acid replication what indicates directly the emerging of newly created virions and their dissemination over the population of non-infected SC. The real significance of the applied DS method could be understood while recognizing it sensitivity to the presence of the particles having certain dipole moments and interacting with the dielectric SC ambient. The general sensitivity of DS is in the range of 0.5–2.5% depending on the instrumentation used. We have shown experimentally that the DS method could be applied for the quantitative independent description of the virus–cell interaction at various stages beginning from the infection emerging for various types of virus–cell pairs due to its general physical modeling. It could be applied both in drug design tests, anti-viral therapy trials as well as in clinical and laboratory practice.

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59
Placental and Monocyte-derived Macrophages have Different Secretome

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It is well documented that placental macrophages (PM) show lower levels of HIV-1 infection than monocyte-derived macrophages (MDM) and that soluble factors secreted in the placenta can inhibit viral replication. We tested the hypothesis that HIV-1 inhibitory factors are secreted by PM and differentially expressed in PM and MDM using proteomics. Cells were cultured for 12 days and supernatant was collected. To characterize PM supernatants, the protein profiles of PM were compared to MDM using the protein chip assay (Ciphergen). The weak cationic exchange (CM10) and metal affinity (IMAC30) surfaces provided the greatest number of protein peaks. Subsequently, proteins were separated by 1D SDS-PAGE and identified by LC–MS/MS. Significant differences were found in four protein peaks with *m/z* values of 6075, 6227, 11,662, 14,547, between PM and MDM supernatants on the CM10 chip and in three protein peaks with *m/z* values of 6158, 7740, 11,934 on the IMAC chip confirming our hypothesis. Proteins were sequenced and identified with high confidence. Worth noting are four peaks that were identified as over-expressed in PM corresponded to fatty acid binding protein-3 (FAB) (14,858 Da) corresponding to the 14,547 *m/z* SELDI-TOF protein peak. Also FKBP 12 (11,951 Da) corresponding with 11,934 *m/z* protein peak, thioredoxin (11,737 Da) and closely correlates with the 11662 *m/z* protein peak. Apolipoprotein E (ApoE, 7558 Da) corresponding to 7740 *m/z* peak. FAB plays a role in transport of fatty acids; FKBP 12 is a peptidil cis–trans isomerase's which aids in the folding of proteins and binds with high affinity to the V3 loop of the HIV-1 envelope glycoprotein; thioredoxin is and antioxidant molecule that could help control viral infection by reducing oxidative stress in macrophages; ApoE mediates binding, internalization and catabolism of lipoprotein particles, and can reduce viral entry. The identity of these proteins found increased in the PM secretome will be validated by Western blots as they could play a role in the inhibition of HIV-1 infection observed in PM.

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60
Inhibition of Human Rhinovirus Replication by Some Antivirals

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Human rhinoviruses, members of the Picornaviridae family, are among the main causative agents of viral upper respiratory tract infections and particularly of common cold. These infections are often mild and self-limiting, but multiple during the human life. The frequent appearance of HRV infections and their economic importance in terms of employee absenteeism, physician visits, and medication costs makes them a subject of primary importance. HRV infections can also be associated with more serious medical complications like acute otitis media, sinusitis, pneumonia and bronchiolitis in infants and young children. Rhinoviral infections commonly cause exacerbations of disease in individuals with underlying respiratory disorders. Until now there is no registered clinically effective antiviral chemotherapeutic agent for treatment of diseases caused by HRVs. Apart from the symptomatic therapy, the hope for an effective treatment of these diseases is the development of broad spectrum antirhinoviral drugs. The topic of the present study is antirhinovirus effect of several picornavirus replication inhibitors with different mode of action against the replication of human rhinoviruses. Monolayer cultures of human cervical carcinoma (HeLa Ohio-I) cells in 96-well tissue culture plates were used in the viral CPE-inhibition test. The action of the compounds at various viral inoculation doses (100, 1000 and 10 000 $CCID_{50}$) was studied to quantitate the antiviral activity and the cytotoxicity of the compounds. The neutral red uptake assay was used. The following compounds have been tested: ribavirin (a large-spectrum viral inhibitor, mostly of RNA viruses), arildone, disoxaril, S7, PTU-23, HBB and oxoglaucine (a compound efficient against enteroviruses initially characterized in our laboratory). The effect of the combinations of most active compounds has been studied. Two of the tested compounds, HBB and oxoglaucine, show the highest activity with a selectivity ratio (CC_{50}/IC_{50}) exceeding 137 and 190, respectively, against 100 cell culture infectious doses 50. According the activity of the tested compounds they can be arranged