

During the last two years of observation the molecular-genetic research allowed to reveal circulation of the two Far Eastern *CA10* genetic lines of different origins and identify the time to their MRCA.

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DISTRIBUTION OF ROTAVIRUS G-, P-, I-, AND E-GENOTYPES IN NIZHNY NOVGOROD, RUSSIAT.A. Sashina¹, O.V. Morozova^{1,2}, N.V. Epifanova¹, T.A. Migunova², N.A. Polyakov², N.A. Novikova^{1,2}¹*I.N. Blokhina Research Institute for Epidemiology and Microbiology, Nizhny Novgorod, Russia, ²Lobachevsky State University, Nizhny Novgorod, Russia*

Rotavirus infection is an important health problem all over the world. In Russia, under the conditions of the beginning of vaccination against this infection, knowledge about its pathogen is limited by the characteristic with the binary classification (G[P]-genotypes), based on the properties of the VP4 and VP7 genes encoding the rotavirus outer capsid proteins. Information about the other gene segments genotypes, as well as unusual and reassortant strains is not sufficient. The aim of this study was to determine the I (VP6) and E (NSP4) genotypes of rotaviruses detected in Nizhny Novgorod using the multiplex PCR method.

We used 55 rotavirus-positive fecal samples from children hospitalized with acute intestinal infection from January to May 2018. RNA of rotaviruses was extracted using "RIBO-prep" reagent kit (AmpliSens, Russia). RT-PCR was carried out with reagents manufactured by "Sileks" (Germany). G- and P-genotypes of rotaviruses were determined using previously published primers. To identify I- and E genotypes in multiplex PCR, fragments of 195 bp (I3), 273 bp (I1), 368 bp (I2) and 233 bp (E3), 305 bp (E2), 443 bp (E1), respectively, were amplified and detected by agarose gel electrophoresis.

I- and E-genotypes were determined in 51 samples (92.8%). In one sample only E-genotype (1.8%) was revealed, and in three — only I-genotype (5.4%). Mostly, the genotypes were detected in combination I1-E1 (52.7%). The set of I2-E2 was found in 30.9% of cases. In addition, the genotype I1-E2 (5.6%) was identified in three samples, I2-E1 and I3-E3 (3.6% together) were shown to be sporadic. The following combinations of G-, P-, I-, and E-genotypes were determined: G1-P[8]-I1-E1 (9.1%), G4-P[8]-I1-E1 (7.3%), G9-P[8]-I1-E1 (32.7%), G4-P[8]-I1-E2 (5.5%), G3-P[x]-I2-E2 (1.8%), G2-P[4]-I2-E2 (29.1%), G2-P[4]-I2-E1 (1.8%), G2-P[4]-I2-Ex (3.6%), G9-P[8]-I1-Ex (1.8%), Gx-P[8]-I1-E1 (5.5%), and Gx-P[x]-I3-E3 (1.8%).

Thus, the new method to identify the I- and E-genotypes was tested and their distribution was determined. Various combinations of G-, P-, I-, and E-genotypes of rotaviruses have been shown. The genotype G9-P[8]-I1-E1 was predominant (32.7%). The G4-P[8]-I1-E2, G3-P[x]-I2-E2, G2-P[4]-I2-E1 strains had probably a reassortant origin.

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DIAGNOSIS OF CYTOMEGALOVIRUS AND PARVOVIRUS B19 INFECTIONS IN SPECIAL GROUPS OF PATIENTS

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Some researchers described the reactivation of cytomegalovirus infection in immunocompetent patients with sepsis, burns, blood transfusions, massive surgical interven-

tions, prolonged mechanical ventilation, use of steroids and vasopressors. In addition to herpesviruses, the reactivation of other latent viruses, in particular, parvovirus B19 (B19V), can also occur with developing immunodeficiency phenomena. With the existing concomitant pathology, these viruses significantly burdens the condition of patients.

For this reason the need for a qualitative and timely diagnosis of viral infections is increasing. PCR assay which capable of detecting even a few molecules of DNA is a progressive diagnostic method due to its high sensitivity. In this regard, quantitative detection of viral DNA can serve as a reliable criteria for significant activity of the pathogen, proving its etiological role in the development of a clinical syndromes.

The aim of the study was to create a test systems for quantitative DNA detection of CMV and B19V with hybridization-fluorescent detection of amplification products in the "real time" mode. It will help to establish the frequency of reactivation of latent viral DNA in critical condition and subsequently determine its effect on the course of the pathological process.

As a result of the studies for the first time in the Republic of Belarus a test systems for the qualitative and quantitative detection of CMV and B19V DNA by the real-time PCR method was created and registered by the Ministry of Health. The main characteristics of the developed test systems showed high values of analytical sensitivity (≥ 2 copies per run of 500 ME/ml), analytical and diagnostic specificity (100%), linear range (> 8 logarithms).

The created test systems, in addition to its use as a diagnostic tool, also can be used as a prognostic marker of infection, as a therapeutic marker for monitoring the success of antiviral therapy as well as for assessing the contagious nature of biological fluids. Thus, during the conducted studies using the test system, reactivation of CMV was detected in 28.6% (6 of 21) of patients in a critical condition with a viral load of 10 to 111 copies/ml. Also, a strong correlation between reactivation of CMV and established diagnosis of sepsis was found ($r = 0.73$). Reactivation of B19V was not detected in any of the 15 patients, which is inconsistent with the existing literature data and requires further researches.

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IMPROVEMENT OF TECHNOLOGY OF PRODUCTION OF HERPETIC VACCINE, CULTURAL, INACTIVE

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To improve methods of production and control of the vaccine, with the aim of developing a new innovative form of herpetic vaccine.

Vaccine strains of herpes simplex virus (HSV) type I (strain "US") and type II (strain "VN") are used as a seed material for preparing of herpesvirus vaccine. The monolayer cell culture (CC) of the primary fibroblasts of chick embryos (FECH) and the diploid cells of the human lung embryo (FLECH) were used for preparing of vaccine. Harvest virus of HSV-I and II types are collected in semi-finished products, which after freezing and thawing are inactivated with formalin. In a comparative plan, the semi-finished products accumulated on different cellular substrates are monitored, in accordance with the production schedule and the current regulatory documents. Semi-finished products are controlled for infectious activity, safety, toxicity, and absence of extraneous contamination. Control of specific activity is carried out in experiments on white rats.