in 2017. Study of susceptibility of *E. coli* showed that resis-
tant to antibiotics strains prevailed (61.8%) and the ratio of
such strains increased almost two-fold in 2017 (72.1%) from 2016 (43.2%). Most isolates were resistant to ampicil-
in and inhibitor-protected penicillins such as amoxicillin/
clavulanate (61.9%). Proportion of such strains increased
almost two-fold in 2017 (72.2%) from 2016 (40.9%). Resistant to
fluoroquinolones, cephalosporins were 3.2% and 2.4% strains, more than 20 times less. All of them were
isolated in 2016. Isolates resistant to two preparations at
the same time were most frequent (58.5%). The proportion of
multidrug resistant cultures was low (2.4%). There were
only 3 isolates resistant to ampicillin, inhibitor-protected
penicillins, fluoroquinolones and cephalosporins.

Number of strains of diarrheagenic *E. coli* increased
two-fold in 2017 compared with 2016, most of them were
*EIEC O 1 44* and *ETEC O25*. Antibiotic-resistant cultures
prevailed. Multidrug resistant strains were rare. All strains
were susceptible to carbapenems, most of them — to flu-
oroquinolones and cephalosporins.

As a result, an algorithm for bioinformational analysis
of outbreaks (cases) of infectious diseases, including those
caused by new (atypical) genetic variants of pathogens of
especially dangerous infections.

**9.16**


**DIFFERENTIATION OF KLEBSIELLA spp. STRAINS
FOR SENSITIVITY TO ANTIBIOTICS USING MASS
SPECTROMETRY ANALYSIS MALDI-TOF**

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*Klebsiella* spp. strains are frequent causative agents
of health care–related infections. Those strains especially
dangerous if they circulate in hospitals. They are usually
antibiotic-resistant. Therefore, information about the sens-
itivity of the isolated strain is necessary in the shortest
time for proper etiological treatment.

The aim of our study was to assess the possibility of using
mass spectrometry analysis (MALDI-TOF) for the rapid
prediction of a selected *Klebsiella* spp. strain resistance.

The study used 195 strains of *Klebsiella* spp. isolated in
various medical centers of St. Petersburg. All strains
were identified by MALDI-TOF. Antibiotic sensitivity was
studied by the disco-diffusion method in accordance with
the recommendations of EUCAST 8.0. We used 19 antibi-
otics from 5 classes for testing strains: aminoglycosides,
beta-lactams, beta-lactams of extended spectrum, quino-
lones and carbapenems.

A hierarchical clustering of spectra was made using the
Unweighted Pair Group Method with Arithmetic Mean
(UPGMA) to determine the relationship between clusters. We used Pearson correlation coefficient between
variable values of peak intensity in spectral profiles as
a measure of the distance between individual mass spect-
tra. We identified 2 significant difference in the spectrum
of the cluster. One cluster included spectra of strains resis-
tant to all studied classes of antibiotics, second cluster —
strains sensitive to them (Distance Level — 0.65). It was
also found that all strains included in the clusters, which
differ from others by Distance Level more than 0.2, have
the same profile of antibiotic resistance.

As a result of this work, we have formed profiles of phe-
notypic resistance of *Klebsiella* spp. strains to 19 antibiotics
and 5 classes. The prospect of using the results of the study
is a significant reduction in the study of biological material
from the patient. Thus, simultaneously with the identifica-
tion of *Klebsiella* spp. strains by MALDI-TOF it is possible
to predict the sensitivity of the isolated strain to different
classes of antibiotics or even to one of them. This will al-
low timely recognition of resistant strains of *Klebsiella* spp
and prescribe adequate etiological therapy, which will sig-
nificantly improve the quality of treatment of patients and
will prevent the spread of resistant strains of bacteria in the
medical establishments.

**9.17**


**PATHOGENIC POTENTIAL OF COMMENSAL
ESCHERICHIA COLI ISOLATED FROM ADULTS
IN SAINT PETERSBURG**

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*Escherichia coli* is one constitute a component of the
natural microbiota of warm-blooded animals including
humans. At the same time commensal *E.coli* is a dynam-
ic population and in some cases is capable to cause ex-
traintestinal diseases, sometimes leading to morbidity and mortality. The aim of this work was to study of genetic diversity and assess of pathogenic potential of commensal *E. coli* isolated from healthy adults in St. Petersburg. 300 *E. coli* strains were collected from fecal samples of 50 St. Petersburg’s inhabitants. *E. coli* strains were isolated using Endo agar and identified by biochemical tests. Determination of four major phylogenetic groups and identification of virulence genes were performed by using real-time, multiplex and simplex PCR. Seven genes typical for ExPEC (*fimH*, *pap*, *aer*, *afa*, *cnf1*, *hlyA*) were identified among the analyzed strains. The B2 phylogroup (47.1%) was leading among other groups: A (20.5%), B1 (9.0%) and D (23.4%). Each strain had at least one virulence gene. No strain had all seven studied genes simultaneously. The maximum number of genes in one strain was five. The prevalence of virulence genes was as follows: *fimH* (98.0%), *pap* (25.0%), *aer* (33.8%), *afa* (5.6%), *cnf1* (11.0%), *hlyA* (10.0%). The strains of groups B2 and D harbored the virulence determinants significantly more frequently than the strains of groups A and B1. Our results showed that *E. coli* isolated from adults differ in their phylogenetic structure and harbour a greater variety of virulence genes. This study revealed that commensal *E. coli* isolated from healthy humans constitute a substantial reservoir of genes related to the extraintestinal pathotypes. All seven tested virulence genes typical for ExPEC were detected and it’s important that the prevalence of these genes was significantly higher among the isolates from healthy adults. So, the extraintestinal virulence genes (encoding the adhesins, toxins, persistence) were found not only in pathogens, but also in commensal microflora of healthy people. Previous reports indicated that virulence genes associated with extraintestinal pathogenesis in fact help the *E. coli* strains to colonize the human gut; therefore, they may be considered as a fitness factor and the virulence is a coincidental side effect.

**9.18**

**EXPRESS METHOD OF GROWING BACTERIA ON THE MEMBRANE OF ANODIC ALUMINIUM OXIDE**

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The classical method of studying bacteria is the cultivation of microorganisms and study of their biological properties. However, this method is very long (several days). So it may not be used in cases when you need to quickly get the result. It can be surgery, sepsis, severe infection, etc. In these cases, the doctor will need a few hours to make a decision on the appointment of causal treatment.

We have developed a method of growing the isolated clones of bacteria from any biological material for 3 hours. The rapid growth of microorganisms is ensured due to the new culture medium. Each microbial cell is grown in a separate cell on a porous membrane of anodic aluminum oxide. After 3 hours of incubation reads visual information using a specially developed image sensor zoom. The visual image of the individual microcolony identified to the species created by special computer programs. The probability of coincidence of the results is 90%. With the help of a special counter counts the number of bacteria of each species in the studied sample. This is especially important in the study of biological material containing several types of microorganisms.

Thus, 3 hours after inoculation of biological material, we get the result about of species and quantitative composition of bacteria. A living culture of microorganisms can work with it further to explore other biological properties, including rapid determination of sensitivity to antibiotics.

**9.19**

**THE DRUG RESISTANCE MUTATIONS OF THE HEPATITIS B VIRUS AMONG HIV-INFECTED INDIVIDUALS**

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Hepatitis B virus (HBV) is one of the most common hepatotropic viruses that can cause both acute and chronic course of the disease. One form of chronic viral hepatitis B is occult hepatitis B, characterized by the presence of HBV DNA in the liver and undetectable levels of HBsAg and HBV DNA in the peripheral blood. The co-infection of HBV with the human immunodeficiency virus (HIV) is facilitated by the common mechanisms and pathways of infection. Although the effect of HBV on the progression of HIV infection appears to be minimal, HIV affects the progression of liver fibrosis, increasing the risk of developing hepatocellular carcinoma and cirrhosis. The need for timely identification HBV variants carrying drug resistance mutations among HBV/HIV-coinfected patients.

The aim of our study was to evaluate the prevalence of HBV with drug resistance mutations among HBV/HIV-coinfected patients.

The material was blood plasma of 264 HIV-infected (HBsAg-) patients with virologic ineffectiveness of ARVT. A method for detecting HBV DNA with a low viral load based on a two-step PCR, followed by sequencing was used. HBV DNA was detected in 89 (33.7%) patients. Based on the phylogenetic analysis it was shown that in this group the HBV subgenotypes are represented in the following ratios: D1 — 39.3%, D2 — 29.2%, D3 — 30.4%, C1 — 1.1%, respectively. In the analysis of nucleotide sequences in the viral polymerase reverse transcriptase domain significant amino acid substitutions (mutations described in the literature as determining the development of drug resistance to nucleotide/nucleoside analogues therapy) were found in 12.35% of patients. Including 9 patient was found to have significant amino-acid replacement in HBV polymerase gene (L180M, M204V) associated with the development of resistance to lamivudine, entecavir, telbivudine and tenofovir. Also in 5.6% of patients were found potentially significant (substitutions in the same significant positions of the polymerase gene, but not described in the literature) — for example L80F.

The obtained data on the prevalence of HBV drug resistance indicate the need for screening of patients with HBV/HIV-coinfected before starting the antiviral therapy.