POSSIBILITIES OF NEW GENERATION SEQUENCING TECHNOLOGY FOR THE DETERMINATION OF RESISTANCE OF *M. TUBERCULOSIS* STRAINS TO ANTIMYCOBACTERIAL DRUGS

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Abstract

The aim of the study was to analyze the possibilities and informativeness of the new generation sequencing technology in the variant of targeted sequencing of the panel of genes responsible for drug resistance to a wide range of antimycobacterial drugs with Deeplex Myc-TB analytical solution for use in clinical practice.

Materials and methods. M. tuberculosis DNA was isolated from 22 cultures obtained on Levenstein-Jensen medium using the QIAamp® DNA Mini Kit. The Deeplex Myc-TB kit was used to amplify the target panel. Sequencing was performed on the Illumina platform (MiSeq, NextSeq 500, Nextera XT library kit). 15 Ukrainian isolates from patients with pulmonary tuberculosis were also characterized by ETR-VNTR PCR typing methods by ETR loci (Exact Tandem Repeats) A, B, C, D and E.

Results. According to the sequence data, out of 15 Ukrainian isolates (from Kyiv city population) *M. tuberculosis* 9 (60.0%) isolates were multidrug resistant and 4 (27.0%) isolates were characterized by broad drug resistance. All MDR and BDR strains had identical mutations, determining resistance to rifampicin (*rpoB1* S450L) and isoniazid (*katG* S315T). Three strains additionally had common mutations that caused resistance to ethionamide (*fabG* C-15T), ethambutol (*embB* Y334H) and streptomycin (*rpsL* K88R). Another group of strains had common mutations in the genes, responcible for resistance to ethambutol (*embB* M306V) and streptomycin (*rpsL* K43R). This may indicate the existence of clusters of strains that are already resistant to these 5 or 4 drugs and prevalent in the population. New mutations with unknown effect on resistance have been found in genes and regions responsible for resistance to rifampicin, pyrazinamide, ethambutol, streptomycin, fluoroquinolones, kanamycin, amikacin, capreomycin, ethionamide.

Conclusions. The Deeplex Myc-TB analytical solution provides a wealth of relevant information on antimycobacterial drug resistance markers and speeds up the data analysis process. To address epidemiological issues, it will be important to use additional typing methods and search for new phylogenetic markers of the most common in the population of clusters of *M. tuberculosis* strains.

Key words: tuberculosis, mycobacteria tuberculosis, drug resistance, polymerase chain reaction, sequencing.

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