networks and change-point analysis reveal key community changes associated with cystic fibrosis pulmonary exacerbations. NPJ Biofilms Microbiomes 2019;5:4.
29. Carmody LA, Caverly LJ, Foster BK, et al. Fluctuations in airway bacterial communities associated with clinical states and disease stages in cystic fibrosis. PLoS One 2018;13(03):e0194060.
30. Hisert KB, Heltshe SL, Pope C, et al. Restoring cystic fibrosis transmembrane conductance regulator function reduces airway bacteria andinflammation in peoplewith cysticfibrosis and chronic lung infections. Am J Respir Crit CareMed 2017;195(12):1617–1628.

**УДК: 616.981.21/.958.7**

# *MICROBIOLOGICAL FEATURES OF THE COURSE OF WIDESPREAD DRUG-RESISTANT PULMONARY TUBERCULOSIS*

Sayfutdinov Z.A.
Center for the development of professional qualifications of medical workers, Tashkent

*ХУЛОСА*

***Тадқиқот мақсади.*** *XDR/BDR-ўпка сили билан оғриган беморларда бактериологик хусусиятлар, бактерия ажралиши даражаси ва қўшимча дорига чидамлилик спектрларини ўрганиш.*

***Тадқиқот материаллари ва усуллари.*** *XDR-туберкулёз ташҳиси қўйилган 135 бемор клиник ва лаборатор жиҳатдан баҳоланди. Балғам намунасининг люминесцент микроскопияси, қаттиқ ва суюқ муҳитларда экиш усуллари, дори сезгирлигини аниқлаш, молекуляр-генетик таҳлиллар амалга оширилди. Тадқиқотга 18 ёшдан 65 ёшгача бўлган 68 нафар эркак (45,8%) ва 147 нафар аёл (67%) киритилган.*

***Натижалар.*** *Беморларнинг кўп қисмида кам миқдорда бактерия ажралиши ва нисбатан тор дорига чидамлилик спектри (BDR+1-3 препарат) кузатилди. Илгари даволанган ва рецидив билан келган беморларда эса кўп миқдорда бактерия ажралиши ҳамда дорига кенг резистентлик (BDR+3-4 препарат) устунлик қилди. Энг кўп учрайдиган резистентлик этамбутол, пиразинамид, ПАСК ва протионамидга нисбатан аниқланди. Беш препаратдан иборат самарали даволаш схемасини танлаш имконияти янги беморларда 63,3%, илгари даволанганларда 31% ва рецидивларда 40% ҳолларда сақланиб қолди.*

***Хулоса.*** *Илгари даволанган XDR/BDR-ўпка сили билан оғриган беморларда бактериологик кўрсаткичлар анча оғир кечади, бу консерватив даволаш самарадорлигини чеклайди ва комплекс ёндашувни, шу жумладан жарроҳлик усулларини қўллашни талаб этади.*

***Калит сўзлар:*** *микробиологик хусусиятлар, дорига чидамлилик, ўпка туберкулёзи.*

*РЕЗЮМЕ*

***Цель исследования.*** *Изучить бактериологические особенности, степень бактериовыделения и спектры дополнительной лекарственной устойчивости у пациентов с XDR/BDR-туберкулезом легких.*

***Материалы и методы исследования.*** *Обследованы 135 пациентов с XDR-туберкулёзом. Проводились люминесцентная микроскопия, посевы на плотные и жидкие среды, определение лекарственной чувствительности и молекулярно-генетические исследования. В исследование вошли 68 мужчин (45,8%) и 147 женщин (67%) в возрасте от 18 до 65 лет.*

***Результаты.*** *У пациентов преимущественно наблюдалось скудное бактериовыделение и относительно узкие спектры лекарственной устойчивости (BDR+1-3 препарата), тогда как у ранее леченных и при рецидивах преобладали массивное бактериовыделение и широкие спектры резистентности (BDR+3-4 препарата). Наиболее частой была устойчивость к этамбутолу, пиразинамиду, ПАСК и протионамиду. Возможность подбора эффективной схемы из пяти препаратов сохранялась в 63,3% случаев у новых пациентов, 31% – у ранее леченных и 40% – при рецидивах.*

***Заключение.*** *У ранее леченных пациентов с XDR/BDR-туберкулёзом отмечаются значительно более тяжёлые бактериологические показатели, что ограничивает эффективность консервативной терапии и требует применения комплексных, включая хирургические, методов лечения.*

***Ключевые слова:*** *микробиологические особенности, лекарственная устойчивость, туберкулёз лёгких.*

World and domestic statistics clearly indicate an increase in the problem of tuberculosis with multiple (MDR) and broad drug resistance (BDR) of mycobacterium tuberculosis (MBT) [6]. Official Russian statistics show that during 2019-2020, the share of MDR–TB patients among newly diagnosed cases and all bacterial isolators with respiratory tuberculosis continued to rise, reaching 32.9-34.0% and 59.9–64.5%, respectively.

In the Russian Federation, the proportion of BDR-TB cases accounts for 18.9% of all reported MDR-TB cases, ranging from 4.4 to 10.2% among newly diagnosed MDR-TB patients, 7.4–13.3% among MDR-TB relapses, and 18.5–30.8% in patients resuming MDR-TB treatment [2]. At the same time, an analytical review of main anti-tuberculosis indicators for 2018–2019 demonstrates an increase in the number of BDR MBT cases registered for treatment (prior to chemotherapy) from 4,775 to 5,347 individuals. In proportion, this corresponds to 36.3% of newly diagnosed cases and recurrent MDR MBT treatment cases. Data on tuberculosis caused by BDR pathogens have long been missing from both domestic and international literature. Nevertheless, BDR-MBT infection is known to follow fundamentally different epidemiological patterns in terms of spread and accumulation within nature and society. It has been established that the elimination of BDR-TB patients from the epidemiological cohort is much slower, unlike patients with other drug resistance spectra of the pathogen, especially drug-sensitive tuberculosis [4].

In pulmonary tuberculosis, the level of bacterial excretion provides an indirect measure of both the cavitary changes in lung tissue and the density of the mycobacterial population. Bronchial patency strongly influences not only the presence but also the extent of excretion. Fluctuations in bacterial levels often reflect how effective chemotherapy has been, yet the heterogeneity of the mycobacterial population can produce so-called "false positive dynamics" [1]. Evidence indicates that even in BDR-TB, certain MBTs located within cavities remain sensitive to specific chemotherapeutic agents or their lower critical concentrations [5], and these are typically eliminated first, leading to an apparent decline in bacterial excretion.

A phenomenon described in phthisiological literature as "rise and fall" occurs when sputum tests, initially negative after cessation of bacterial excretion, later return positive as drug-resistant pathogen populations accumulate [7]. Observing such patterns in BDR-TB is crucial because they signal inadequate chemotherapy effectiveness. Furthermore, "false positive" bacteriological results can obscure latent drug resistance, potentially delaying necessary adjustments in chemotherapy regimens or decisions regarding surgical intervention. Despite these limitations, assessing the magnitude of bacterial excretion remains an essential component of evaluating the overall clinical picture of the disease [3].

## THE AIM OF THE STUDY

To investigate the bacteriological characteristics, degree of bacterial excretion, and patterns of additional drug resistance in patients with XDR/BDR pulmonary tuberculosis.

## MATERIAL AND METHODS

A prospective study was carried out on 135 patients with extensively drug-resistant (XDR) pulmonary tuberculosis. The cohort included 68 men (45.8%) and 147 women (67%) aged between 18 and 65 years. The examination of sputum included luminescent microscopy, cultivation on both solid and liquid nutrient media for subsequent assessment of drug susceptibility, as well as molecular genetic techniques to identify MBT and determine the genetic markers associated with pathogen drug resistance. Sputum cultures were performed on liquid media using the automated BACTEC 960 MGIT system, with determination of sensitivity to first-line anti-tuberculosis drugs; and on solid Löwenstein–Jensen media, followed by transfer to a medium for MBT drug sensitivity (DS) testing via the absolute concentration method. Bacterial excretion was assessed using both the seeding method and luminescent microscopy. Cultures were considered sensitive to critical drug concentrations if fewer than 20 colonies appeared in a test tube that showed abundant growth in the control.

Critical concentrations were set as follows: rifampicin – 40 mcg/ml, isoniazid – 10 mcg/ml and 1 mcg/ml, streptomycin – 10 mcg/ml, ethionamide/protionamide – 30 mcg/ml, kanamycin – 30 mcg/ml, cycloserine – 30 mcg/ml, capreomycin – 30 mcg/ml, ethambutol – 2 mcg/ml, and ofloxacin – 4 mcg/ml. Using the seeding method, bacterial massiveness was classified as abundant (over 100 CFU), moderate (21–100 CFU), or meager (1–20 CFU). In luminescent microscopy, the categories were abundant (more than 10 ARM per field), moderate (1–10 ARM per field), and meager (10–99 ARM per 100 fields of view).

The cessation of bacterial excretion was recorded from the month in which both bacterioscopy and seeding first yielded negative results, provided that subsequent tests remained negative. Analyses of the pathogen's drug resistance (DR) spectra and resistance to individual chemotherapy drugs across different patient groups identified a total of 22 distinct MBT DR spectrum variants. Statistical processing was performed using Statsoft STATISTICA 10 and Microsoft Excel 2016. Correlations between treatment safety indicators and the frequency of adverse events were evaluated using rank-based nonparametric Spearman coefficients.

## RESULTS AND DISCUSSION

The results of assessing the degree of bacterial excretion according to the method of luminescent microscopy and seeding on liquid nutrient media are presented in tab. 1.

Table 1 shows that low bacterial excretion was mainly seen in newly diagnosed patients, with luminescent microscopy indicating 18 individuals (72%) and the seeding method identifying 16 patients (64%). By contrast, massive bacterial excretion occurred significantly more often in patients receiving repeated courses of chemotherapy – 73 individuals (73%) by luminescent microscopy and 70 patients (70%) by the seeding method.

In patients experiencing disease relapses, massive excretion was observed even more frequently, with 6 individuals (60%) by luminescent microscopy and 8 patients (80%) by the seeding method.

*Table 1*

**The massiveness of bacterial excretion in XDR/TB patients of different registration groups**

| Group of patients | Number of patients | Luminal microscopy of sputum | | Sputum sowing | |
|---|---|---|---|---|---|
| | | MBT massiveness (+) | | MBT massiveness (+) | |
| | | Meager | Massive | Meager | Massive |
| Newly identified | 25 | 18 | 7 | 16 | 9 |
| % | | 72% a b | 28% c d | 64% e f | 36% g h |
| Previously treated | 100 | 28 | 73 | 30 | 70 |
| | | 28% a | 73% c | 30% e | 70% g |
| Relapse | 10 | 4 | 6 | 2 | 8 |
| % | | 40% a | 60% d | 20% f | 80% h |
| Total | 135 | 50 | 85 | 59 | 74 |
| % | 100% | 37%±4,8 | 63%±3,6 | 43,7%±4,5 | 54,8%±4,0 |

Note: a-a; b-b; c-c; d-d; e-e; f-f; g-g; h-h – the difference is statistically significant, p<0.05

Regarding drug resistance, newly diagnosed patients predominantly displayed LU spectra with minimal additional resistance. Specifically, among 23 newly diagnosed patients (92%) harboring a BDR pathogen, additional resistance was limited to a maximum of three chemotherapy drugs. In contrast, patients undergoing repeated treatment exhibited broader MBT resistance spectra. Additional resistance to the 2nd, 3rd and 4th drugs during repeated treatment was noted, respectively, in 18 (18%), 30 (30%) and 30 (30%) patients. In patients with relapses, the leading position was occupied by a spectrum of drugs with additional resistance to 3 CP – 3 patients (30%). The most frequently observed combinations among all patients were as follows: BDR+3 CP in 40 patients (29.6%), BDR+4 CP in 32 patients (23.7%), and BDR+2 CP in 27 patients (20%).

The diagram in Fig. 1 clearly illustrates the spectra of additional drug resistance in BDR-TB patients across the different registration groups.

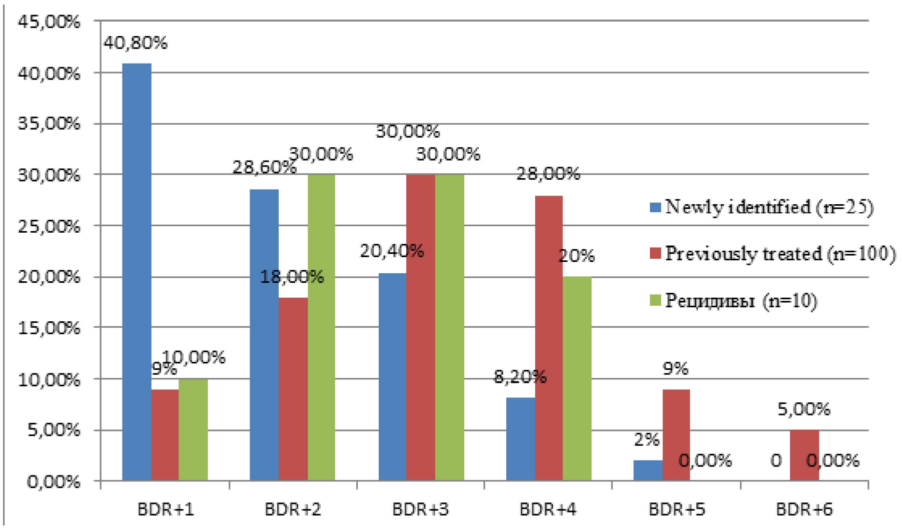


*Fig. 1.* Spectra of additional DR in BDR-TB patients of different registration groups.

The data presented in the diagram in Fig. 1 indicate that among newly diagnosed patients, BDR+1 combinations were significantly more prevalent compared to other groups (p<0.05). In contrast, BDR+2, BDR+3, and BDR+4 combinations were significantly more frequent in previously treated and relapsed patients compared to newly diagnosed patients (p<0.05).

Drug resistance across different patient categories is presented in Fig. 2. Among previously treated patients, resistance was most commonly observed to pyrazinamide (75%), followed by ethambutol (70%), PASC (55%), and protionamide (48%). In newly diagnosed patients, on the other hand, the spectra of additional resistance were dominated by protionamide (38.8%), pyrazinamide (38.8%), ethambutol (40.8%), and PASC (30.6%). Among relapsed patients, the highest resistance rates were recorded for pyrazinamide (60%), ethambutol (60%), PASC (60%), and protionamide (52.4%). Resistance to fourth-generation fluoroquinolones was detected in 40.8% of newly diagnosed patients, 50% of previously treated patients, and 50% of relapsed patients.

Calculations indicate that the identified spectra of additional drug resistance of the BDR pathogen allow for a combination of five chemotherapy drugs with preserved pathogen sensitivity in 63.3% of newly diagnosed patients, 31% of previously treated patients, and 40% of patients with recurrent TB.

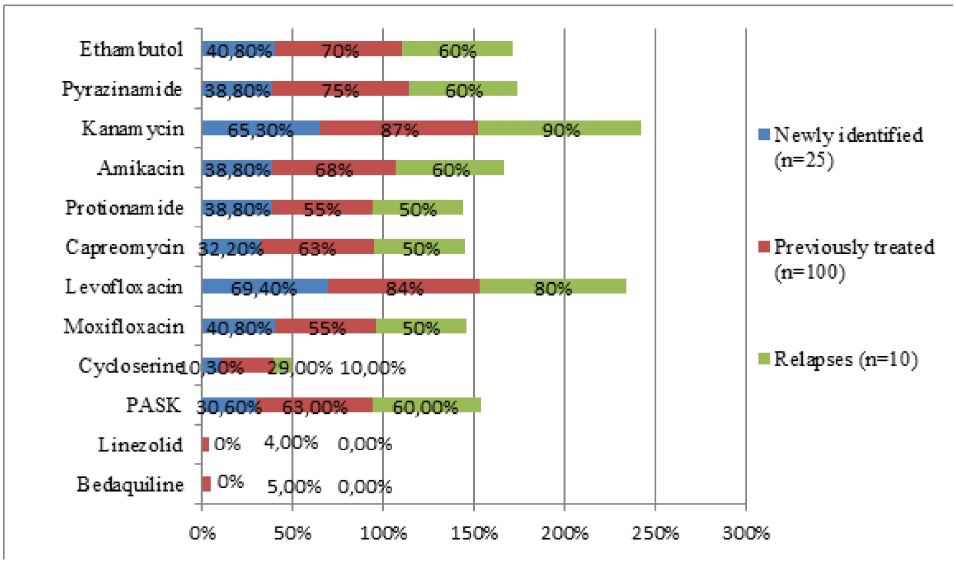


*Fig. 2.* Additional drug resistance of BDR-MBT to individual CP in patients in different registration groups.

CONCLUSION

In patients with previously treated pulmonary BDR-TB, bacteriological features were markedly more pronounced across most parameters, which considerably restricted the effectiveness of conservative treatments. As a result, the overall prognosis of the disease was largely determined by access to additional complex therapeutic approaches, including surgical procedures.

REFERENCES


1. Abdullaev, R.Yu. Hepatotoxic reactions in the treatment of newly diagnosed patients with pulmonary tuberculosis with multidrug–resistant pathogen / R. Yu. Abdullaev, O. G. Komissarova, E. S. Chumakova [et al.] // Tuberculosis and lung diseases. – 2019. – Vol. 97. – No. 7. – P. 21-27. (In Russian)
2. Baranova, O.N. Pulmonary tuberculosis with multidrug–resistant pathogen // Notes of a scientist. – 2015. – No. 6-1. – P. 19-21. (In Russian)
3. Kuzmina, N.V. The course and effectiveness of treatment of tuberculosis with multiple and broad drug resistance // Fundamental and applied problems of human health in the North: A collection of materials of the V All-Russian Scientific and practical conference, Surgut, October 27, 2020. Surgut: Surgut State University. – 2020. – P. 82-88. (In Russian)
4. Kranzer, K. New WHO Treatment Recommendations for Multidrug-Resistant Tuberculosis: Are We Well Enough Prepared? // Am J Respir Crit Care Med. – 2019. – Apr 26. [Epub ahead of print].
5. Vjecha, M.J. Accelerating the development of therapeutic strategies for drug-resistant tuberculosis // Nat Rev Drug Discov. – 2018. – Vol.17(9). – P.607-608.
6. World Health Organization. WHO consolidated guidelines on drug-resistant tuberculosis treatment. Geneva, World Health Organization. – 2020.
7. Zhao, Y. Improved treatment outcomes with bedaquiline when substituted for second-line injectable agents in multidrug resistant tuberculosis: a retrospective cohort study // Clin Infect Dis. – 2018.


-------------------------------------------------------------