Clinical and Immunological Characteristics of Drug-Resistant Tuberculosis in Children and Adolescents

Mordovskaia L.I., Gur'eva O.I.

ABSTRACT

We studied characteristics of clinical progression and immunological indicators in children and adolescents with drug-resistant tuberculosis. Control group consisted of patients with drug-sensitive tuberculosis. It was concluded that drug resistance of M.tuberculosis adversely influences the clinical progression and results in markedly suppressed immune system indicators in children and adolescents.

Keywords: child, adolescent, drug-resistant tuberculosis, immune system.

INTRODUCTION

Extremely difficult epidemiologic situation for tuberculosis (TB) in general and pediatric populations is still existent in Russia [1,4]. TB incidence rate in children and adolescents is one of the most sensitive indicators of the epidemiologic situation. Under existing epidemiologic situation, pulmonary TB is present in two variants, with different clinical symptoms, progression and outcomes, and thoroughly different approaches to multimodality treatment: drug-sensitive (DS) pulmonary TB, cause by *M.tuberculosis* (MTB) with sensitivity to all first and second line drugs; drug-resistant (DR) pulmonary TB, caused by MTB with resistance to one, two or more drugs/drug combinations. There is a separate entity of multidrug-resistance (MDR), which is characterized by simultaneous resistance to isoniazid and rifampicin regardless of resistance to other drugs. Due to age-related specifics, the problem of DR TB, as well as MDR TB, in children and adolescents, needs a relevant approach to disease detection and treatment. Dependence between the disease progression pattern and MTB drug-resistance pattern is not excluded, as far as studies by Russian researchers report [2,3].

Aim: To study clinical progression and immunological indicators specific for pediatric DR TB.

MATERIAL AND METHODS

Clinical radiological findings and immunological data of 24 pediatric patients with DR TB and 19 with DS TB were comparatively studied. At the time of study (2009-2013), all children and adolescents were undergoing in-patient therapy at the Pediatric Department no.2, Research & Practice Center for TB ('Ftiziatriia'), Sakha Republic (Yakutia).

Clinical, bacteriological, immunological, x-ray and CT examinations were performed in all patients. During physical examination, attention was paid to presence of the following signs and symptoms: general intoxication symptoms (body weight loss, loss of appetite, pallor, low grade fever, fatigue, sweating). Mantoux skin test with 2 TU PPD-L was performed in compliance with the instruction on the use of tuberculin skin tests given in the Russian Federation Health Ministry Order no. 109 (dated 2003). Skin tests using Diaskintest (recombinant TB allergen in standard dilution (0.2mcg), 0.1 ml solution for intradermal administration) were performed in accordance with labeling.

Bacteriologic tests included luminescence microscopy, culture on solid and liquid media (BACTEC MGIT 960). For immunological analysis, we studied relative lymphocyte count and lymphocyte subset composition in peripheral blood ($CD3^+$, $CD4^+$, $CD8^+$, $CD20^+$). Immunophenotyping was done using immunofluorescence method with appropriate monoclonal antibodies produced by Becton Dickinson (USA). Immunofluorescence response was assessed using flow FACSCalibur cytofluorometer (Becton Dickinson, USA). Serum cytokine profile was determined using ELISA test system produced by ZAO Vektor-Best (Russia). Whole blood IFN-gamma induction was studied in the presence of MTB antigens (tuberculin, ESAT-6 /CFP-10), using Tubinferon test-kits (Russia).

RESULTS

Age-sex distribution of patients is shown in Table 1.

As is shown in Table 1, adolescence age group predominated. There were more girls than boys among patients with DR TB (16 (66.7%) vs. 8 (33.3%), respectfully). There was no difference in numbers of girls and boys in the group of patients with DS TB (9 (47.4%) vs. 10 (52.6%), respectfully).

Positivity for MTB was detected by culture in all 43 patients (100.0%). Acid-fast bacteria (AFB) were detected by microscopy in 15 (62.5%) patients with DR TB and in 8 (42.1%) with DS TB. MDR MTB were detected in 16 cases with DR TB (66.7%).

Study of DR spectrum in patients with DR TB (Fig. 1) showed that resistance developed more often to isoniazid (100%), streptomycin (94.5%) or rifampicin (88.9%). DR to kanamycin (27.7%), etambutol (16.7%) and capreomycin (16.7%) was less common. In adolescents, DR spectrum was identical to the infection source DR spectrum in 17 (70.8%) of cases. Contact with active TB was established in 20 (83.3%) patients with DR TB, intrafamilial contact occurred in all patients. Among patients with DS TB, contact with active TB was detected in 10 (52.6%) patients, 8 (42.1%) of them were intrafamilial, and moreover, MTB of the infection sources were sensitive to all drugs.

Of the patients with DR TB, 19 (79.2%) had marked symptoms of TB intoxication. In patients with DS TB, the majority had moderate (9 (47.4%)) or mild (y 7 (36.8%)) TB intoxication symptoms.

Laboratory tests showed marked alterations in blood pictures of patients with DR TB: heightened ESR in 24 (100%), leucocytosis in 18 (75%), and lymphopenia in 15 (62.5%) (Table 2).

Results of tuberculin skin tests with 2 TU PPD-L did not differ much between the patients with DR or DS TB. Mean papule sizes for Mantoux skin test with 2 TU PPD-L and Diaskintest are show n in Table 3.

As is seen from Table 3, mean papule size after skin test with 2 TU PPD-L was 17.3±0.6 mm. (DR TB) and 16.5±1.0 mm. (DS TB) ($p>0.05$). Mean papule sizes after Diaskintest were 12.5±0.5 mm. (DR TB) and 12.4±1.0 mm. (DS TB) ($p>0.05$).

Analysis of clinical forms of TB showed the presence of infiltrative pulmonary TB in the majority of patients: 20 (83.2%) cases in DR TB and 14 (73.7%) cases in DS TB ($p<0.001$). (Fig. 2).

Fig. 2 shows that patients with DR disease had besides infiltrative lung TB the following diagnoses: nodular (focal) lung TB (2; 8.4%); caseous pneumonia (1; 4.2%); TB of the bronchi (1; 4.2%). Patients with DS disease were diagnosed, apart from infiltrative lung TB, with primary complex (3 15.7%), caseous pneumonia (1; 5.3%) and nodular (focal) lung TB (1; 5.3%).

Analysis of the x-ray findings showed that extensive (bilateral) pulmonary TB was seen most often in patient s with DR (13; 54.2%) versus DS (3; 15.8%) cases ($p<0.01$). Lung cavities were observed more often in DR TB, than in DS TB: 15 (62.5%) cases versus 7 (36.8%) cases ($p<0.01$).

The results of the study of the immune status in patients with DR TB are shown in Table 4.

As is seen in Table 4, patients who isolated DR MTB had marked alterations in the T-cell component of the immune system: decreased $CD3^+$ counts in DR (51.3±0.9%) vs. DS (56.8±2.1%) TB ($p<0.05$); decreased $CD4^+$ counts in patients with DR TB (36.3±1.1%; $p<0.001$), decreased $CD4^+/CD8^+$ ratio (mean decrease rate was 1.2; $p<0.05$). $CD20^+$ counts were elevated in all patients and no meaningful differences were noticed between patients isolating DS or DR TB. Levels of $CD16^+$ were 17.3±1.1% in DR group and 20.7±1.2% in DS group ($p<0.05$).

Levels of PPD-induced IFN-gamma production were 189.4±47.8 mg/mL in patients with RD TB and were higher (247.0±64.9 pg/mL) in patients with DS TB. The same difference was

true for blood specimens incubated in the presence of ESAT-6/CFP-10 antigens: levels of IFN-gamma in DR and DS TB were 74.0±17.6 pg/mL and 136.8±31.8 pg/mL, respectively ($p<0.05$). Levels of antigen-induced IFN-gamma in whole blood of patients with pulmonary TB were inversely related to the disease extent and severity. Serum IFN-gamma production in patients with DR TB showed a clear declining trend, compared to patients with DS TB. IL-8 counts in patients with DR TB were elevated up to 71.0±36.8 pg/mL (as compared to 12.6±4.5 pg/mL in patients with DS TB). IL-4 content tended to increase in patients with DR TB.

CONCLUSION

Drug resistance of MTB is a factor that negatively influences TB disease progression in pediatric patients. DR TB in children and adolescents is associated with marked symptoms of tubercular intoxication, hyperergic reaction to Mantoux skin test with 2 TU PPD-L, destruction of lung tissue, and stable decline of the immune status indicators.

References:

1. Aksenova V.A. Tuberkulez u detei i podrostkov v Rossii (Problemy i puti ikh resheniia) [Tuberculosis in children and adolescents in Russia (Problems and solutions)]. In: Sb. materialov nauch.-prakt. konf. “Tuberkulez u detei i podrostkov” [Abstract book of the research & practice conference ‘Tuberculosis in children and adolescents‘]. Moscow; 2009. p. 22-25. Russian.

2. Poluektova F.G. Osobennosti techeniia i effektivnost’ lecheniia lekarstvenno-ustoichivogo tuberkuleza legkikh u podrostkov [Disease progression and treatment effectiveness in adolescents with drug-resistant pulmonary tuberculosis]. Synopsis of Cand.Med.Sc.(PhD) Thesis. Moscow; 2004. Russian.

3. Firsova V.A. et al. Techenie tuberkuleza legkikh u podrostkov v zavisimosti ot raznoi stepeni lekarstvennoi rezistentnosti [Pulmonary tuberculosis in adolescents in relation to different degree of drug resistance]. Problemy Tuberkuleza, 2002, no. 12, pp. 23-25. Russian.

4. Shilova M.V. Tuberkulez v Rossii v 2008 godu [Tuberculosis in Russia in 2008]. Moscow; 2009. Russian.

Authors:

Mordovskaia Larisa Ivanovna, Dr.Med.Sc. (MD), senior researcher, Head of the Immunological Laboratory, Research & Practice Center for Tuberculosis, mobile: +7(914)272-42-69, email: limordovskaya@mail.ru

Gur'eva Olga Ivanovna, Cand.Med.Sc. (PhD), Head of the Pediatric Department no.2, Research & Practice Center for Tuberculosis, mobile: +7(924)176-93-54, email: gurievaolga@mail.ru