
Diagnosis of Tuberculosis Infection Activity by ELISA and Transcription Analysis Methods

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Abstract: Enzyme immunoassay is unable to detect the degree of development of the disease. In addition, antibodies to tuberculosis in the body do not in all cases indicate that the patient is infected. This method allows you to diagnose tuberculosis of the joints. The study showed a low level of diagnostic value of P. Mantu 2TE in the diagnosis of tuberculosis in children infected with MBT, which requires the introduction of new methods. In the examined children with low sensitivity to tuberculin in 21.8% of cases and with average sensitivity in 40.7% of cases, the activity of tuberculosis infection detected during the Diaskin test is determined, which is accompanied by a reaction of VGLU according to the results of radiation examination in 14.3% and 53.0% of cases, respectively. The study showed a low level of diagnostic value of P. Mantu 2. Diagnosis of tuberculosis in children infected with MBT, which requires the introduction of new methods. In the examined children with low sensitivity to tuberculin in 21.8% of cases and with average sensitivity in 40.7% of cases, the activity of tuberculosis infection detected during Diaskintest is determined, which is accompanied by a reaction of the values according to the results of radiation examination in 14.3% and 53.0% of cases, respectively.

Keywords: tuberculosis, Diaskin test, Enzyme immunoassay, computed tomography, diagnostics.

Introduction. ELISA is an antibody test. This method helps to establish the presence of immunoglobulin in relation to the Koch stick. It is often used as an alternative study for the Mantoux reaction. Traditionally, the diagnosis of tuberculosis of VGLU consists of the results of a clinical examination of children, tuberculin diagnostics (P. Mantoux 2TE, performing a graded skin test [HCP]) and a standard X-ray examination, including an overview X-ray and linear chest tomography. Thus, the low specificity of the Mantoux test dictates the need to introduce new immunological tests for early detection and determination of the activity of tuberculosis infection in children. In recent years, along with the Mantoux test, WHO recommends the use of the Quantiferon test, T-spot test. Preliminary results of its use indicate that the Diaskin test is highly informative; however, comparative studies on the effectiveness of the use of new immunological tests in the diagnosis of tuberculosis in children have not been conducted. Moreover, it is of particular interest to compare the results obtained with traditional methods - P. Mantu 2 T E, in order to form new approaches to the

Diagnosis of tuberculosis. Diaskintest (Tuberculosis recombinant allergen)-the domestic drug, which is used only in Russia, has been used since 2010. It contains two antigens present in virulent strains of *Mycobacterium tuberculosis* and absent in the BCG vaccine. Diaskintest is administered in the same way as the Mantoux test - intradermally. Evaluation of the skin reaction at the injection site is also carried out after 72 hours. Unlike tuberculin, there are only 2 antigens in Diaskintest, so it has a higher sensitivity and specificity than the Mantoux test, that is, it is more likely to show a reaction to tuberculosis, if there is one, and less likely to give a false positive reaction.

Materials and methods. For the period from 2019-2021, a prospective study was conducted in the Department of Pediatric phthisiology to study the informativeness of Diaskintest in the complex diagnosis of tuberculosis in children. 119 younger children (from 3 to 7 years 49 people [41.7; $m = 4.5 \pm 0.2$]) and older were examined (from 7 to 13 years – 70 [58.3; $m = 12.3 \pm 0.3$]) of age. The analysis of the results of the tuberculin Mantoux 2TE skin test was carried out by introducing purified tuberculin in standard dilution (ready-made form), and a sample with a tuberculosis recombinant allergen in standard dilution - Diaskin test (DST). Quantitative determination of interferon gamma (IFN γ) by enzyme immunoassay

(ELISA, ELISA) was used to detect in vitro cellular response to stimulation by these peptide antigens associated with Mycobacterium tuberculosis infection. In 66.8% (78) of cases, patients were identified and referred for examination on increasing sensitivity to tuberculin, 35 people were examined by contact with a tuberculosis patient (29.2%), and 5 children by complaint (4.1%). According to the anamnesis, all children included in the study were vaccinated with BCG in the maternity hospital, 65 (54.2%) of them were effective, as evidenced by a post-vaccination scar of more than 4 mm. Manifestations of intoxication syndrome were absent in 21 (17.5%) children, were moderate in 47 (39.2%) and pronounced in 52 children (43.3%). They served as the basis for radiation examination (MSCT and MSCT angiography). The assessment of the condition of the intra-thoracic lymph nodes in children during radiation diagnosis in our work was carried out in accordance with the recommendations according to which, with a transverse size of the lymph node in children from 3 to 14 years of age more than 5 mm, it is necessary to interpret these changes against the background of MBT infection as "small" manifestations of intra-thoracic adenopathy. The analysis of the diagnostic accuracy of the applied tests and methods is carried out by calculating operational characteristics: diagnostic sensitivity (DH), diagnostic specificity (DS), predictive value of positive (PCR) and negative results (PCR), as well as diagnostic effectiveness (DE).

Results and discussion

Intoxication syndrome was moderate in 39.1% (46) cases, pronounced in 43.1% (51), and absent in 16.5% (20). At the same time, patients had numeric sensitivity to tuberculin according to Mantoux 2TE in 62.5% (75; $m = 12.3 \pm 0.2$) of cases, in 17.5% (21) there was a low level of specific sensitization ($m = 5.5 \pm 0.2$), in 20.0% (24) hypermetric sensitivity. According to DST, the test was negative in 58 (48.3) people, doubtful in 6 (5.0) ($m = 3.2 \pm 0.1$) and in 56 (46.7) (12.7; $m = 18.0 \pm 0.3$) children positive. The CF test in 57.5% (69) showed a negative result, in 41.7% (50) – positive, in 0.8% (1) – doubtful, which in 87.5% confirms the positive result of DST and allows us to determine a differentiated approach to the appointment of CF. In the complex of serological reactions, the titer of specific antibodies for all reactions was lower than diagnostic in 56.7% (68) of cases, in 30.8% (37) – positive for one and two reactions, which is possible in the presence of MBT infection. In 12.5% (15) of cases in children, a positive AT titer was determined by three and four reactions, which indicated the activity of tuberculosis infection.

A direct dependence of the severity of intoxication syndrome was revealed with negative and positive Diaskin-test, as well as with positive and negative CF. The absence of clinical symptoms was noted significantly often with negative DST (66.7) and CF (80.9), with positive DST (67.3) and CF (59.6), the symptoms of intoxication were significantly often pronounced. There was no similar dependence in children with different sensitivity to tuberculin according to Mantoux 2TE and manifestations of intoxication syndrome. The analysis of the results of immunological tests (Diaskin-test, P. Mantoux 2TE) and radiological (MSCT and MSCT-AG) characteristics of intra-thoracic lymph nodes in the

examined children was carried out. In children with low and medium sensitivity to tuberculin, positive results were noted. Attention was drawn to the absence of positive tests in 20.8% for DST and 25.0% for CF in patients with high sensitivity to tuberculin, which may indicate a false positive reaction to tuberculin. In 25.0%

Of cases, according to the results of radiation examination, patients with high sensitivity to tuberculin according to MSCS have no reaction of VGLU. The comparison of the results of Diaskintest and CF test in the examined children showed almost identical data. The predominance of negative results on CF with a negative DST of 98.3 was noted. % and positive CF with

a positive DST of 87.5%. Thus, CF in 95% of cases confirms the results of DST, which gives reason to consider their diagnostic value comparable. The reaction of VGLU during radiation examination (MSCT and MSCT angiography) in patients with the absence of activity of tuberculosis infection according to the results of DST (negative) was absent in 43.1% in 31.1% of cases it was visualized up to 0.5 cm in size, which is the norm. With a positive DST, the reaction of LU is more

0.5 cm was significantly more common – in 80.4%, in 73.9% of cases - with lime deposition. The results obtained allowed us to confirm the opinion about the critical size of the VGLU of more than 0.5 cm during the radiation complex examination, since it was precisely this reaction of the LU that was reliably often encountered against the background of positive DST and CF. In children with a positive Diaskintest, pronounced symptoms of intoxication were noted in 67.3% of cases, and 80.4% were visualized LU is more than 0.5 cm according to MSCT and CT angiography, in 73.9% of cases - in the phases of reverse development, which confirms the presence of tuberculosis infection activity. The data obtained allow us to recommend a radiation examination in children with positive DST, as a risk group for tuberculosis, in order to reasonably prescribe adequate therapy. Indicators of the diagnostic value of the Diskin test (DCH - 77.3, DS - 89.4, PCPR - 91.1,

PCOR - 72.4, DE - 77.5) in comparison with P. Mantu 2TE (DCH - 84.1, DS - 19.6, PCPR - 53.2, PCOR - 47.6, DE - 57.2) is twice as high, which proves

It's high in formativeness and the need to use it in a mandatory complex of phthisiological examination.

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