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Our Experience in Diagnosing Tuberculous Spondylitis

Objective — to increase the efficiency of diagnosis in patients with tuberculous spondylitis (TS).

Materials and methods. The clinical material of this research consisted of protocols of clinical and radiological examination, and analysis of the results of the study of 30 patients with active TS (the main group). Criteria for inclusion of patients in the study: TS in the active phase (verified pathomorphologically and/or bacteriologically). The diagnostic group included 20 patients who were also referred to our hospitals with suspected TS, but the diagnosis was not confirmed during an in-depth examination. The average age of patients in the main and diagnostic groups was (38.2 ± 9.6) and (40 ± 10.8) years, respectively ($p > 0.05$).

Results and discussion. In total, we examined 45 patients from both groups who were admitted to hospital with suspected spondylitis using surgical biopsy (30 patients in the main group and 15 in the diagnostic group). Surgical biopsy diagnostic methods included abscess puncture, puncture and trepanation biopsy of the vertebral bodies. Punctures of paravertebral abscesses were performed in 21 patients. Puncture and trepanation biopsy of the vertebral bodies was used in 24 patients. Through biopsy study using surgical techniques, the diagnosis of TS in the main group was confirmed in 30 patients. Bacteriological confirmation of the diagnosis was obtained by us in 12 (39.96 %) patients: in 6 (19.98 %) patients *Mycobacterium tuberculosis* (MTB) was isolated only by the culture method, in the others only by the bacterioscopy method and in 2 (6.72 %) cases positive results were obtained by both methods. The diagnosis of a spinal tumour was established in 4 patients in the diagnostic group; another 4 patients from this group were diagnosed with non-specific spondylitis, and 7 with osteochondrosis of the spine was identified. In no case of puncture and trepanation biopsy did we observe any complications. DNA of the MTB complex was detected by polymerase chain reaction (PCR) in 40 (88.8 %) of 45 samples of surgical material from patients with a histologically confirmed diagnosis of TS. Overall, confirmation of the diagnosis of TS by PCR and culture methods was obtained in 93.7 % of cases.

Conclusions. Assessment of the results of bacteriological examination of patients showed a relatively low efficiency of bacteriological diagnostic methods for TS and a high efficiency of PCR. Biopsy findings indicate the high efficiency of this method and its in conducting aetiological diagnosis with mandatory testing of the sensitivity of MTB to antibacterial drugs. Improving the diagnosis of TS is an important factor in achieving better treatment outcomes for this disease.

Keywords

Differential diagnosis of TS, biopsy, PCR, bacteriological diagnosis.

Tuberculous spondylitis (TS) is fundamentally characterised by the destruction of the bone tissue of the vertebral bodies or their processes, secondary involvement of the intervertebral discs, formation of a soft tissue component of inflammation in the paravertebral soft tissues and epidural space, with development of inflow abscesses [1, 3, 4].

Diagnostic difficulties in specific spinal lesions are a generally recognized fact. Diagnostic errors in this disorder range from 40 to 75 %. The reason for this is the polymorphism of specific osteomyelitis with various forms of course, similar in clinical and radiological presentation to a number of musculoskeletal diseases and prolonged uncon-

trolled intake of various antibacterial agents before diagnosis [2, 11, 12].

One of the main conditions for successful treatment of TS is its timely diagnosis before the development of such formidable complications as neurological deficit, suppurative abscesses, fistulas, spinal deformity etc. [2, 9, 10].

All diagnostic methods in TS can be divided into two main types: detection of characteristic changes in the tissues of the musculoskeletal system and detection of the tuberculosis pathogen. There are direct and indirect diagnostic methods respectively. Detection of characteristic changes in the tissues of the musculoskeletal system involves direct morphological and radiological methods, indirect ones are classical methods of patient examination, laboratory studies and functional diagnosis. Detection and identification of the pathogen imply direct diagnostic methods such as microscopy, culture isolation and molecular diagnosis, and indirect methods such as tuberculin diagnosis, antigen determination and determination of anti-tuberculosis antibodies by enzyme-linked immunosorbent assay (ELISA) [4, 6, 7, 11].

The development of a number of new technologies for radiological diagnosis of the spine has significantly expanded diagnostic capabilities. Contrast myelography, computed tomography, magnetic resonance imaging and ultrasound diagnosis have been added to the traditional radiological method. New high-tech methods have raised hopes for accomplishment of all diagnostic tasks. Accumulated world experience and our data show that none of the above methods, when used in isolation, solves all diagnostic problems [2, 9, 12].

Diagnosis of TS is based on a comparison of clinical, laboratory, bacteriological and radiological data, and new methods of radiological diagnosis. It is also necessary to take into account the nature of the disease, usually gradual and prolonged, previous or concomitant tuberculosis lesions of other organs or systems, and the presence of conditions that contribute to contact with tuberculosis patients.

Currently, in the usual clinical and radiological presentation of TS, there are deviations: isolated lesions of one or more vertebrae, the posterior supporting complex, multiple lesions of the spine at different levels. In some cases, the bone process develops against the background of anti-tuberculosis therapy, which is carried out for a different localisation of the infection, which changes its radiological manifestations and time course. Spondylitis often develops secondary to dystrophic changes, especially in elderly patients. These features, as well as the growth of purulent, oncological, dystrophic and other related abnormalities of the skeleton, cause difficulties in the diagnosis and differentiation of TS at the present time [5, 8, 11].

Difficulties in early detection lead to a relatively late diagnosis of the disease, already during the development of the destructive process of the adjacent surfaces of the vertebrae and the occurrence of neurological complications and abscesses.

Diagnostic errors that occur at the early stages of examination of patients with spinal tuberculosis play a leading role in the occurrence of advanced forms of tuberculous spondylitis leading to disability [10, 12].

It should be noted that most often differential diagnostic difficulties are noted when distinguishing TS from congenital and acquired degenerative-dystrophic changes in the spine, primary and metastatic tumours and non-specific osteomyelitis of the spine.

Objective — to increase the efficiency of diagnosis in patients with tuberculous spondylitis.

Materials and methods

The clinical material for this research included protocols of clinical and radiological examination, analysis of the results of the examination and treatment of 30 patients (the main group) with active tuberculous spondylitis, who were treated and examined on beds for patients with bone and joint tuberculosis in the pulmonary and surgical department of the Regional Anti-Tuberculosis Dispensary No. 1 (RATD) of Kharkiv and the clinical base of the Department of Traumatology and Orthopaedics of Kharkiv National Medical University (KhNMU) in the departments of traumatology and orthopaedics of the Municipal Non-profit Enterprise Professor O.I. Meshchaninov City Clinical Hospital of Emergency and Urgent Medical Care of Kharkiv City Council in the period from 2015 to 2021, where patients with TS were treated who did not pose any threat to others in epidemiological terms, the so-called «closed» forms of the disease (without the presence of fistulas, connecting tuberculosis lesions of the lungs).

Criteria for inclusion of patients in the study: TS in the active phase (verified pathomorphologically and/or bacteriologically).

Criteria for exclusion of patients from the study: age older than 75 years; decompensated concomitant (non-tuberculosis) lesions.

In the majority of cases, lesions of two vertebral bodies were noted: 25 (83.25 %). The thoracolumbar spine was most often affected. It should be noted that when the destructive specific process was localised in these parts of the spine, the involvement of three vertebral bodies in the inflammatory process was more often observed.

The diagnostic group included 20 patients who were also referred to the above-mentioned hospitals

with suspicion of TS, but the diagnosis was not confirmed during an in-depth examination.

The average age of patients in the main and diagnostic groups was (38.2 ± 9.6) and (40 ± 10.8) years respectively ($p > 0.05$).

Aetiological diagnosis of destructive spinal lesions

The generally accepted gold standard for establishing the nature of destructive spinal lesions is pathomorphological and bacteriological methods. These research methods require the collection of pathological material for assessment directly from the focus of destruction or from inflowing abscesses or fistulas that are directly connected with the area of pathological lesions.

The most highly informative and often the only reliable methods for diagnosing the inflammatory process in the spine are those based on the study of tissue diagnostic material. Various methods of surgically obtaining biopsy material in our hospitals were used after a complex of clinical, radiological and laboratory studies.

Methods of bacteriological study

Microscopic study is known to be the most accessible and rapid method for determining *Mycobacterium tuberculosis* (MTB). The most frequently used bacteriological methods for the detection of MTB are direct microscopy of smears and microscopy of smears prepared after enrichment (centrifugation, flotation, microflotation) with subsequent staining by the Ziehl—Neelsen method or with luminescent dyes.

Bacteriological studies and polymerase chain reaction (PCR) were performed in the bacteriological laboratory of RATD No. 1 in Kharkiv together with a bacteriologist.

Collection and preparation of pathological material for the study were performed under sterile conditions. The punctate of the inflow abscess obtained with a syringe and the pathological material taken during biopsy of the destruction focus were placed in sterile dishes and immediately delivered to the laboratory.

In fistulous forms of tuberculosis, discharge from the fistula was taken for bacteriological analysis. In case of abundant discharge, pus was collected directly into a test tube. In case of scanty pus discharge, the fistula was washed with sterile isotonic sodium chloride solution, and the washings collected in a test tube were sent for examination. Due to the frequent oligobacillary nature, as well as the reduced viability and enzymatic activity of MTB isolated from the foci, a wide range of nutrient media used for the isolation of MTB was employed for cultural diagnosis. The growth of the first colonies on classical media was noted after 4—8 weeks.

Study by polymerase chain reaction method

For the research, we used the certified PCR test system Ampli-Sens-100 MTB-com. The research itself and the interpretation of the results were performed according to the instructions (protocol) in the certified laboratory of RATD No. 1 of the city of Kharkiv.

The choice of clinical material for the study is determined by the most likely location of the pathogen. It is fundamentally important that the pathological material being studied is in contact with the focus of destruction; in our case, it is pus from suppurative abscesses or fistula material.

The amount of material for the study should not be excessive, since together with the pathogen, substances that can cause inhibition of PCR or can contribute to DNA degradation during storage and transportation get into the sample.

To perform PCR, it is necessary to take 1—2 ml of the punctate of the suppurative abscess into a dry sterile tube; the material for the study must be fresh, not cooled and not frozen.

Further examination was performed according to the study protocol.

Surgical and puncture methods for obtaining biopsy material

In total, we examined 45 patients from both groups who were admitted to the hospital with suspected spondylitis using surgical biopsy (of which 30 patients were the main group and 15 were in the diagnostic group).

Biological material obtained from the destruction foci studied during the study included pus, granulations, sequestrations, fragments of intervertebral discs and bones.

Surgical biopsy diagnostic methods included abscess puncture, puncture and trepanation biopsy of the vertebral bodies. Punctures of paravertebral abscesses were performed in 21 patients. Puncture and trepanation biopsy of the vertebral bodies was used in 24 patients. It was carried out under the control of an electron-optical converter (EOC). Puncture of retroperitoneal abscesses was also carried out under ultrasound control.

Pathomorphological studies

Pathomorphological examinations of the surgical material obtained from patients in both groups were carried out on the basis of the pathological and anatomical department of RATD No. 1 in Kharkiv. In complex cases, advisory assistance was provided by the laboratory of connective tissue morphology and experimental modelling of the State Institution Professor M.I. Sytenko Institute of Spine and Joint Pathology of the National Academy of Medical Sciences of Ukraine.

For histological examination of the surgical material of patients (bone sequesters, necrotic areas of bone, granulations, pyogenic membranes of suppurative abscesses etc.), the pathological material was fixed in 10 % formalin solution. Then the soft tissues were embedded in paraffin, and the bone tissue was decalcified in 5 % nitric acid solution and embedded in celloidin. The sections were stained with haematoxylin-eosin and picrofuchsin according to Van Gieson and light-optical examination was performed.

Results and discussion

Bacteriological confirmation of the diagnosis was obtained in 12 (39.96 %) patients, of whom 6 (19.98 %) patients had MTB isolated only by the culture method, in the others only by the bacterioscopy method, and in 2 (6.72 %) cases, positive results were obtained by both methods.

Analysis of the informativeness of various pathological material showed that the largest number of positive results, both by the culture method and by bacterioscopic method, was obtained from the contents of abscesses in 10 (33.33 %) patients, pus on tampons and discharge from fistulas had significantly fewer findings in 2 (6.66 %) patients.

Thus, bacteriological studies confirm the data of other authors that a feature of all pathological material in TS is its oligobacillary nature [3, 4]. This requires a special attitude to microbiological examination, great care in its conduct and the need to improve bacteriological methods for diagnosing TS.

Results of biopsy studies

As noted above, we examined a total of 45 patients who were admitted to the hospital with suspicion of TS using surgical biopsy. Biopsy studies using surgical techniques confirmed the diagnosis of TS in 30 patients in the main group.

Punctures of paravertebral abscesses were performed in 21 patients. A positive result was considered the detection of bacterial flora or MTB in the punctate; however, their absence could not indicate against the diagnosis of TS.

In the absence of an effect during puncture biopsy, the next stage of the surgical diagnostic method was performed, namely percutaneous trephine biopsy of the vertebral bodies. This method was used to diagnose tuberculous spondylitis in 10 patients in the main group. The diagnosis of spinal tumour was established in 4 patients of the diagnostic group, another 4 patients of this group were diagnosed with non-specific spondylitis, and 7 patients of the diagnostic group had spinal osteochondropathy. In 5 patients of this group, the diagnosis of TS was made using modern methods of radiographic imaging (MRI, CT) i. e. these patients

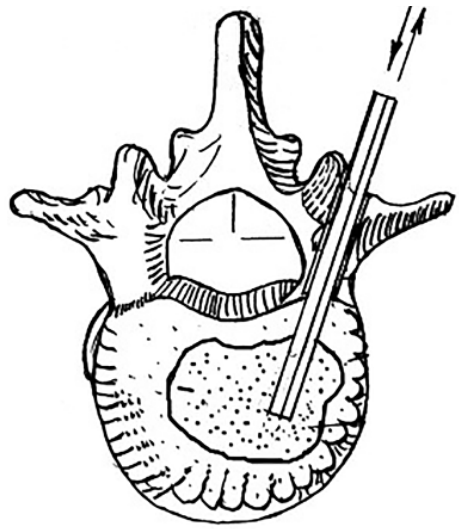

Figure. **Scheme of trepanobiopsy**

did not require biopsy studies. In no case did we have any complications when using puncture and trepanation biopsy. Trepanation biopsy was performed using EOC with posterior access through the root of the arch of the affected segment. The scheme of performing this manipulation is presented in Figure.

Laboratory examinations using surgical methods of obtaining biopsy material before establishing the diagnosis lasted from two days to two weeks.

Thus, the use of surgical methods of obtaining biopsy material in spondylitis and other diseases of the spine in all cases that cause any difficulties in determining the aetiology of the pathological process in the spine allows avoidance of diagnostic errors and significantly reduce the time of diagnostic examination to 2 weeks instead of 2—3 months which are usually spent on diagnostic dynamic observation of patients and obtaining final results of bacteriological studies.

Results of molecular genetic methods of etiological diagnosis

Studies to detect MTB-complex DNA were carried out in 45 samples. Isolation of total DNA from clinical samples was performed using the kit for the isolation of nucleic acids Amplitub-RV M-sorb-tub-2 according to the manufacturer's instructions.

MTB-complex DNA was detected by PCR in 40 (88.8 %) of 45 surgical specimens from patients with histologically confirmed diagnosis of TS. Several pathological specimens were taken from individual patients with TS.

It should be emphasised that the PCR method has a significant advantage in both sensitivity and speed of obtaining the analysis result.

Thus, given the uniqueness of the diagnostic material and the need to obtain a culture of MTB

not only to verify the diagnosis of TS but also to determine the drug sensitivity of strains, a comprehensive approach to choosing methods for isolating the pathogen is necessary.

In total, a positive result, namely the detection of the pathogen by cultural and/or molecular genetic methods, was obtained in the study of 28 patients (93.7 %) of surgical specimens from patients with histologically verified diagnosis of TS.

When comparing diagnostic efficiency, the molecular genetic method showed an undeniable advantage in sensitivity (88.8 %) over bacteriological methods (39.96 %, in total when cultured on dense and liquid nutrient media).

At the same time, a positive result obtained by the culture method, with a negative PCR result, was observed in only one case.

Analysis of different types of surgical material revealed the same diagnostic informativeness of both material containing pus and granulations and material including bone fragments.

Analysis of difficulties and errors in the diagnosis of TS

Most patients in the main group were admitted to the above-mentioned departments after several stages of inpatient treatment in other medical institutions with erroneous diagnoses.

When analysing the medical documentation of these patients, we found that a targeted examination of the spine at previous stages of treatment was often not carried out, despite the increase in pain after symptomatic treatment and the occurrence of spinal deformation. The main reason for the development of significant destructive changes in the spine was the late diagnosis of the disease.

Thus, in 5 patients (16.5 %) an X-ray examination was first performed only after the appearance of signs of spinal cord compression. A serious cause of diagnostic errors was poor-quality X-ray examination of the spine and failure to use such modern methods of radiographic imaging in the diagnosis of TS as CT, MRI, serological and cultural research methods. In 3 patients (9.9 %) the tuberculous process at a relatively early stage was not detected since the spine radiographs were performed at several levels above or below the lesion site. In 2 cases (6.6 %) errors were made due to incorrect interpretation of low-quality radiographs.

In 4 cases, the erroneous diagnosis was due to atypical clinical and radiological manifestations of TS. Reassessment of the diagnostic significance of individual signs of the disease contributed to the fact that, due to the incorrect diagnosis, 2 patients underwent radiotherapy, 4 patients received narcotic analgesics. In 2 cases, surgical interventions (laparotomy, opening of suppurative abscesses, laminectomy) were mistakenly performed without appropriate indications, which contributed to a deterioration of the patients' condition and activation of the tuberculous process.

It should be noted that the difficulties in distinguishing spinal abnormalities have increased significantly in recent decades due to the observed pathomorphosis of TS. Marked ageing of the patient population, changes in the cultural properties of mycobacteria, and widespread use of antibiotics often change the classic course of spinal tuberculosis, giving rise to atypical, clinical and radiological features that may contradict conventional ideas about TS. Similar phenomena of pathomorphosis are also currently observed in cases of non-specific osteomyelitis and oncological conditions. Therefore, an attempt to establish a diagnosis on one radiological or clinical sign, — even one that may appear pathognomonic — was the main cause of erroneous diagnosis in most of our cases.

The most characteristic symptoms that provoked diagnostic errors in the differentiation of tuberculosis and spinal tumours were: pronounced destruction of the body of only one vertebra while the neighboring vertebrae appear intact, which occurs in the modern course of TS, the absence of pronounced convergence of the remains of the bodies of destructively altered vertebrae in the elderly, selective damage to the posterior parts of the spine (posterior spondylitis).

In 9 cases, the cause of the formation of advanced forms of TS was a long asymptomatic clinical course of the tuberculous process in the spine and the associated late referral of patients to a doctor.

When examining patients, we used traditional clinical, radiological and laboratory methods of examination, which are generally available.

Assessment of findings in the patients of the diagnostic group (n = 20) showed that among 12 patients with confirmed degenerative-dystrophic lesions of the spine, in 3 patients a congenital anomaly of the spine development (vertebral synostosis, wedge-shaped vertebra) was mistaken for TS, in 6 osteochondropathy of the vertebral bodies, in 3 hormonal and involutional spondylopathy (at the previous stages of treatment). The diagnosis of a spinal tumour was established in 4 patients and in 4 more patients a diagnosis of non-specific spondylitis was detected (the diagnostic group).

The basis of differential diagnostic errors was the similarity of individual clinical and radiological manifestations of these diseases of the spine, which are different in their origin.

Differentiation of TS from degenerative-dystrophic changes in the spine was complicated not only

by the similarity of the presentation, but also by the absence of clearly defined destructive changes in the vertebral bodies at the initial stage of spondylitis development using conventional X-ray examination methods and the presence of degenerative changes in the adjacent parts of the spine in elderly people. These changes simulated TS by narrowing of the intervertebral spaces and deformation of the bodies of adjacent vertebrae, and the formation of large Schmorl's hernias in the vertebral bodies, which were very similar to destructive foci.

In all 4 patients of the diagnostic group with cancer metastases to the spine, changes in the vertebrae were the only manifestation of the tumour process noted upon admission. Upon further examination, it was determined that in 2 cases the source of metastasis was lung cancer, and in 2 cases prostate cancer.

Tumours simulated tuberculosis by the common manifestations of pain syndrome in the early stages of the disease, subfebrile temperature, the presence in several patients of traces of past pulmonary tuberculosis, deformation of the bodies of two adjacent vertebrae, destructive foci in the vertebral bodies and abscess-like paravertebral shadows formed by tumour-like masses extending beyond the vertebra.

Errors in the differential diagnosis of tuberculous and non-specific osteomyelitis of the spine occurred with endogenous infection of tuberculous foci with secondary microflora and the associated manifestation of such symptoms uncharacteristic of tuberculosis as high fever, sclerosis of the affected vertebrae and the formation of osteophytes. Non-specific osteomyelitis in 4 patients of the diagnostic group, in turn, under the influence of antibacterial therapy, took on the course of the process inherent to tuberculosis and had less pronounced characteristic reactions of the endosteum and periosteum.

It should be noted that the main methods of examination in the differential diagnosis of non-specific osteomyelitis of the spine and TS are bacteriological and pathomorphological studies.

Thus, the study of patients admitted to hospital with a preliminary diagnosis of TS allows us to conclude that there are significant difficulties in the differential diagnosis of this disorder and a high frequency of errors in establishing the diagnosis at an early stage of the disease. In addition, diagnostic errors made by doctors at the early stages of patient examination play a leading role in the development of complicated forms of TS.

The reasons for this lie both in the insufficient diagnostic capabilities of the general medical network due to the lack of necessary equipment and the vigilance of specialists regarding these diseases, and in the changed clinical and radiological picture of TS at the current stage.

In this regard, it is necessary to improve the methods of diagnosis and differential diagnosis of these dangerous diseases, to form risk groups for TS within the general medical network, and to alert doctors who provide primary care to extrapulmonary forms of tuberculosis.

Conclusions

1. Assessment of the results of bacteriological examination of patients showed a relatively low efficiency of bacteriological diagnostic methods for TS and a high efficiency of PCR.

2. Biopsy findings indicate high efficiency of this method and feasibility in conducting aetiological diagnosis with mandatory testing of the sensitivity of MTB to antibacterial drugs.

3. Improving the diagnosis of TS is an important factor in improving the results of treatment of this disease.

There is no conflict of interest.
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Наш досвід діагностики туберкульозного спондиліту

Мета роботи — підвищити ефективність діагностики хворих на туберкульозний спондиліт (ТС).

Матеріали та методи. Клінічним матеріалом були протоколи клініко-рентгенологічного обстеження й аналіз даних 30 пацієнтів з активним ТС (основна група). Критерії залучення пацієнтів у дослідження: ТС в активній фазі (верифікований патоморфологічно та/або бактеріологічно). Діагностична група утворена з 20 пацієнтів, направлених у стаціонарний етап обстеження та лікування з підозрою на ТС, але результати детального обстеження дали змогу заперечити цей діагноз.

Результати та обговорення. Із застосуванням хірургічної біопсії було обстежено 45 хворих обох груп, госпіталізованих у стаціонар із підозрою на спондиліт (з них 30 пацієнтів основної групи та 15 діагностичної групи). Методика хірургічної біопсійної діагностики полягала в пункції абсцесів, пункційній і трепанаційній біопсії тіл хребців. Пункцію паравертебральних абсцесів виконано в 21 хворого. Пункційну і трепанаційну біопсію тіл хребців застосовано в 24 хворих. Біопсійним дослідженням із використанням хірургічних методик діагноз ТС в основній групі був підтверджений у 30 хворих. Бактеріологічне підтвердження діагнозу отримано в 12 (39,96 %) хворих, з них у 6 (19,98 %) *Mycobacterium tuberculosis* (МВТ) виявлено лише методом посіву, у решти — лише методом бактеріоскопії, у 2 (6,72 %) — двома методами. У 4 хворих діагностичної групи діагностовано пухлину хребта, ще в 4 — неспецифічний спондиліт, у 7 — остеохондропатію хребта. У жодному випадку використання пункційної і трепанаційної біопсії не зареєстровано будь-яких ускладнень. ДНК МВТ-complex виявлена методом полімеразної ланцюгової реакції (ПЛР) у 40 (88,8 %) із 45 зразків операційного матеріалу пацієнтів із гістологічно підтвердженим діагнозом ТС. Загалом діагноз ТС установлено за результатами ПЛР і культуральними методами в 93,7 % випадків.

Висновки. Аналіз результатів бактеріологічного дослідження пацієнтів засвідчив порівняно низьку ефективність бактеріологічних методів діагностики при ТС і високу ефективність ПЛР. Результати біопсійних досліджень підтвердили їхню високу ефективність і доцільність використання в етіологічній діагностиці з обов'язковим дослідженням чутливості МВТ до антибактеріальних препаратів. Удосконалення діагностики ТС є важливим чинником поліпшення результатів лікування цього захворювання.

Ключові слова: диференційна діагностика, біопсія, полімеразна ланцюгова реакція, бактеріологічна діагностика.

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