Infection and Drug Resistance

Dovepress
open access to scientific and medical research

Open Access Full Text Article

ORIGINAL RESEARCH

Analysis of the Value of Serum Biomarker LBP in the Diagnosis of Spinal Tuberculosis

Caili Lou[1,2], Jian Liu[2], Zhibo Ren[2], Jinke Ji[2], Hongbao Ma[2], Hui Dong[3], Linan Wang[2], Xu Zhang[3], Ningkui Niu[1]

[1]Department of Spinal Surgery, General Hospital of Ningxia Medical University, Yinchuan, Ningxia, 750004, People's Republic of China; [2]Clinical Medical School, Ningxia Medical University, Yinchuan, Ningxia, 750004, People's Republic of China; [3]Institute of Medical Sciences, General Hospital of Ningxia Medical University, Yinchuan, Ningxia, 750004, People's Republic of China

Correspondence: Ningkui Niu, Department of Spinal Surgery, General Hospital of Ningxia Medical University, No. 804 Shengli Street, Yinchuan, 750004, People's Republic of China, Tel +86 13629593280, Email niuningkui6743242@163.com; Xu Zhang, Institute of Medical Sciences, General Hospital of Ningxia Medical University, No. 804 Shengli Street, Yinchuan, Ningxia, 750004, People's Republic of China, Email xuzhang1012@163.com

Objective: To investigate the correlation between the expression of lipopolysaccharide-binding protein (LBP) in peripheral blood of spinal tuberculosis and clinical diagnosis and to evaluate its value as a diagnostic marker of spinal tuberculosis.

Methods: In the experimental group, clinical history data and peripheral blood were collected from 100 patients with spinal tuberculosis who were admitted to the Department of Spine Surgery, General Hospital of Ningxia Medical University from May 2017 to May 2020, and peripheral blood was collected from 30 healthy volunteers in the control group. Screening of differential LBP expression by proteomics and ELISA to verify its expression in peripheral blood of spinal tuberculosis patients. *t*-test, Spearman analysis, linear regression and ROC curve were used to evaluate the diagnostic value of LBP in peripheral blood for spinal tuberculosis.

Results: The expression of LBP protein in peripheral blood is significantly higher in patients with spinal tuberculosis than in the normal population; LBP assay values were significantly and positively correlated with CRP and ESR values ($P < 0.01$); the AUC of LBP in the diagnosis of spinal tuberculosis for pathological examination, bacteriological culture, T-cell spot test for tuberculosis infection (T-SPOT), imaging diagnosis, and acid fast bacillus were, respectively, 0.677 ($P < 0.01$), 0.707 ($P < 0.01$), 0.751 ($P < 0.01$), 0.714 ($P < 0.01$), and 0.656 ($P < 0.05$), and there was a correlation between LBP and the diagnostic evaluation of spinal tuberculosis.

Conclusion: LBP could be a new candidate biomarker for the diagnosis of spinal tuberculosis.

Keywords: spinal tuberculosis, proteomics, LBP, biomarker

Introduction

Tuberculosis due to infection by the bacterial pathogen Mycobacterium tuberculosis represents one of the ten most potent killers and the deadliest disease due to a single pathogen, more than HIV/AIDS, totaling 5.8 million newly diagnosed cases and approximating 1.3 million deaths in 2020.[1] Spinal tuberculosis (STB) is often secondary to pulmonary tuberculosis and is the most common bone and joint tuberculosis, accounting for approximately 50% of all bone and joint tuberculosis, which predisposes to serious complications such as spinal cord and nerve damage.[2] The early clinical symptoms of STB patients are atypical, the onset is insidious, and the duration of the disease is long, which makes it difficult to confirm the diagnosis and makes it easy to be misdiagnosed and missed. Early diagnosis of the disease can reduce the risk of pain and complications. Therefore, it is of great significance to explore molecular markers with high specificity and sensitivity for the clinical diagnosis and treatment of STB disease. Lipopolysaccharide-binding protein (LBP) is a 60kDa molecular protein that belongs to type I reactive proteins and plays an important significant role in infectious diseases or immune responses.[3] The literature has confirmed that LBP is expressed directly in human peripheral hematogenous monocytes, interacting with human macrophages and monocytes.[4] Its specific and stable expression in cardiovascular and neoplastic diseases is expected to be the potential for detecting molecular markers of diseases.[5–7] However, the potential capabilities of LBP are not yet clear. In

Received: 14 June 2022
Accepted: 15 August 2022
Published: 29 August 2022

this study, we screened and validated the differential expression of LBP as a molecular marker in the peripheral blood of STB patients and healthy subjects by proteomics and analyzed the correlation of its clinical characteristics to evaluate the potential of LBP as a diagnostic biomarker for STB, hoping to provide reference and ideas for the diagnosis and treatment of STB.

Materials and Methods

Clinical Data

From May 2017 to May 2020, the Department of Spine orthopedics of Ningxia Medical University admitted 100 exceptions to the peripheral blood and clinical data of patients with spinal tuberculosis were collected for the experimental group and 30 cases of healthy examinees, of which 50 cases of males and 50 cases of females in the experimental group, aged 18–77 years old, the average age (49.47±16.32 years old), the control group of 13 cases of males, 17 cases of females, age 40–72 years old, the average age (53.39±9.67 years old). There was no statistical significance in the comparison of sex and age in both groups ($P>0.05$).

All experiments involving human blood samples were conducted in accordance with the Declaration of Helsinki. The experimental protocol of this study was reviewed and approved by the Ethics Committee of the General Hospital of Ningxia Medical University.

Inclusion criteria for patients with spinal tuberculosis:[8] (1) Clinical symptoms (such as: night sweats, back pain, fatigue, etc), laboratory and imaging tests are comprehensively diagnosed as patients with spinal tuberculosis; (2) Patients with spinal tuberculosis who have not been treated with anti-tuberculosis before surgery; (3) Patients with spinal tuberculosis who have not recently taken hormones or immunosuppressant drugs.

Criteria for exclusion in patients with spinal tuberculosis:[8] (1) Patients with diabetes, malignant tumors and other infectious diseases; (2) Patients who may cause changes in blood routine values due to recent medication; (3) Women in menstrual cycles; (4) Patients with purulent infections and lumbar spine lesions such as Brucella; (5) Participants with incomplete information or refusal to participate in research and research.

Inclusion criteria for healthy people: (1) Previously healthy, no chronic diseases such as hypertension, no history of diabetes, malignant tumors, tuberculosis, and no immune activating or inhibitory drugs have been taken in the past 1 month; (2) There is no history of viral and bacterial infections in the past two weeks; (3) There is no abnormal performance in physical examination, and routine blood tests show that macrophages, monocytes, granulocytes and other indicators are within the normal range; (4) The reagents and methods used in the blood routine of the experimental group and the control group are consistent.

Criteria for exclusion of healthy people: (1) The control group has active pulmonary tuberculosis, diabetes, malignant tumors and infectious diseases; (2) Patients who may have recent medications that may cause changes in blood routine values; (3) Women in the menstrual cycle; (4) immunodeficiency diseases or other immune system diseases; (5) Participants have incomplete information or refuse to participate in surveys.

Clinical Data Collection of Patients

We collected basic clinical data of STB patients and treatment related to STB diagnosis, including name, sex, age, course of disease, laboratory tests (blood routine, CRP, ESR, etc), bacterial culture, pathological examination, T-SPOT, imaging results and other examination results.

Sample Preparation

All participants received fasting peripheral blood samples in the early morning and were stored in a vacuum anticoagulant tube. Then, the collected peripheral blood samples were centrifuged and packed in the laboratory within 1h. The centrifuge conditions were as follows: at 4°C for 3000 RPM/min for 10 minutes. After completion, the upper serum was taken and placed in a 1.5mLEP tube and stored at −80°C.

Proteomics Detection and Analysis

Three pairs of peripheral blood of STB patients and healthy persons were selected as experimental groups and control groups, and Tandem Mass Tag quantitative proteomics detection was performed using the high-resolution mass spectrometer Q-Exactive plus, and the differential protein data were analyzed using mass spectrometry analysis software Mascot 2.6 and Proteome Discoverer 2.2 after mass spectrometry scanning to determine the unilateral edge of the preliminary screening differences. The preliminary filter criteria are: difference multiples ≥1.5 times, using Cluster 3.1, GraphPad Prism 8.0.1 software to map differential protein clusters and volcanoes.

Gene Ontology (GO) and Kyoto Encyclopedia of Genes and Genomes (KEGG) Signaling Pathway Analyses

Metascope, a web-base resource (http://metascope.org) for gene and protein annotation, visualization, and integration discovery, was used to perform GO analyses. The KEGG (KEGG Orthology And Links Annotation) online analysis database (http://www.kegg.jp/blastkqalal) was used to perform KEGG pathway analyses.

ELISA Validation of LBP Differential Proteins

The relevant ELISA experiments should be started after the collection of the number of samples in the experimental combination control group is completed. We selected 50 STB patients and 30 health examiners for peripheral blood to verify the differences in LBP. The ELISA kit (LBP antibody kit, Beijing Xinbosheng Technology Biotechnology Co, Ltd.) were used to measure LBP concentrations within the range of the standard curve. A standard curve was constructed according to the manufacturer's instructions. The measurement range for the LBP kit was 15–350 pmol/L.

Statistical Methods

SPSS26.0 software and GraphPad Prism Version 8.0.1 software were used for statistical analysis and processing. Measurement data were expressed as X ± S; *t*-test was used to compare the data between the two groups. Curvilinear regression and Spearman test were used to analyze the correlation between LBP and other counts. ROC curve was used to evaluate the specificity and sensitivity of LBP in the diagnosis of spinal tuberculosis. The area under curve (AUC) ≥0.6 was considered to be of diagnostic value. Inspection level $P<0.05$ was considered statistically significant.

Results

Screening and Validation of LBP Proteins

Under the preliminary proteomic screening, a total of 16 differentially expressed proteins were found in STB patients and healthy peripheral blood, of which 11 were upregulated differential proteins and 5 were expressed in STB peripheral blood. According to cluster analysis heat maps, advanced volcano maps show differential protein expression in STB and healthy peripheral blood (Figures 1A and B). Expanded filter criteria: the difference multiple ≥1.5, $P < 0.01$, the associated protein was related to the STB signaling pathway; we used ELISA to verify LBP expression in peripheral blood between STB patients and healthy people, and the results confirmed that LBP does have significant differences in the peripheral blood of STB patients and meets the screening criteria. ELISA verifies that LBP differential protein expression differs between healthy and STB peripheral blood (Figure 1C). The ROC curve shows: LBP specificity and sensitivity in the peripheral blood of STB patients, AUC of 0.880 ($P < 0.01$) indicates that LBP has the potential to detect STB markers (Figure 1D).

Results of LBP Analysis in GO and KEGG

GO and KEGG analyses are important methods and tools in the field of bioinformatics for analyzing the functional enrichment of differentially expressed proteins. Thus, we used these analyses to identify differences between children with vs without MPP. In Table 1, the GO functional enrichment analysis of the STB group versus normal controls included, among others, cellular process of GO, biological regulation, localization, metabolic process and immune response. In Table 2, the KEGG pathway enrichment analysis showed that among others, NF-κB signaling pathway, TLRs signaling

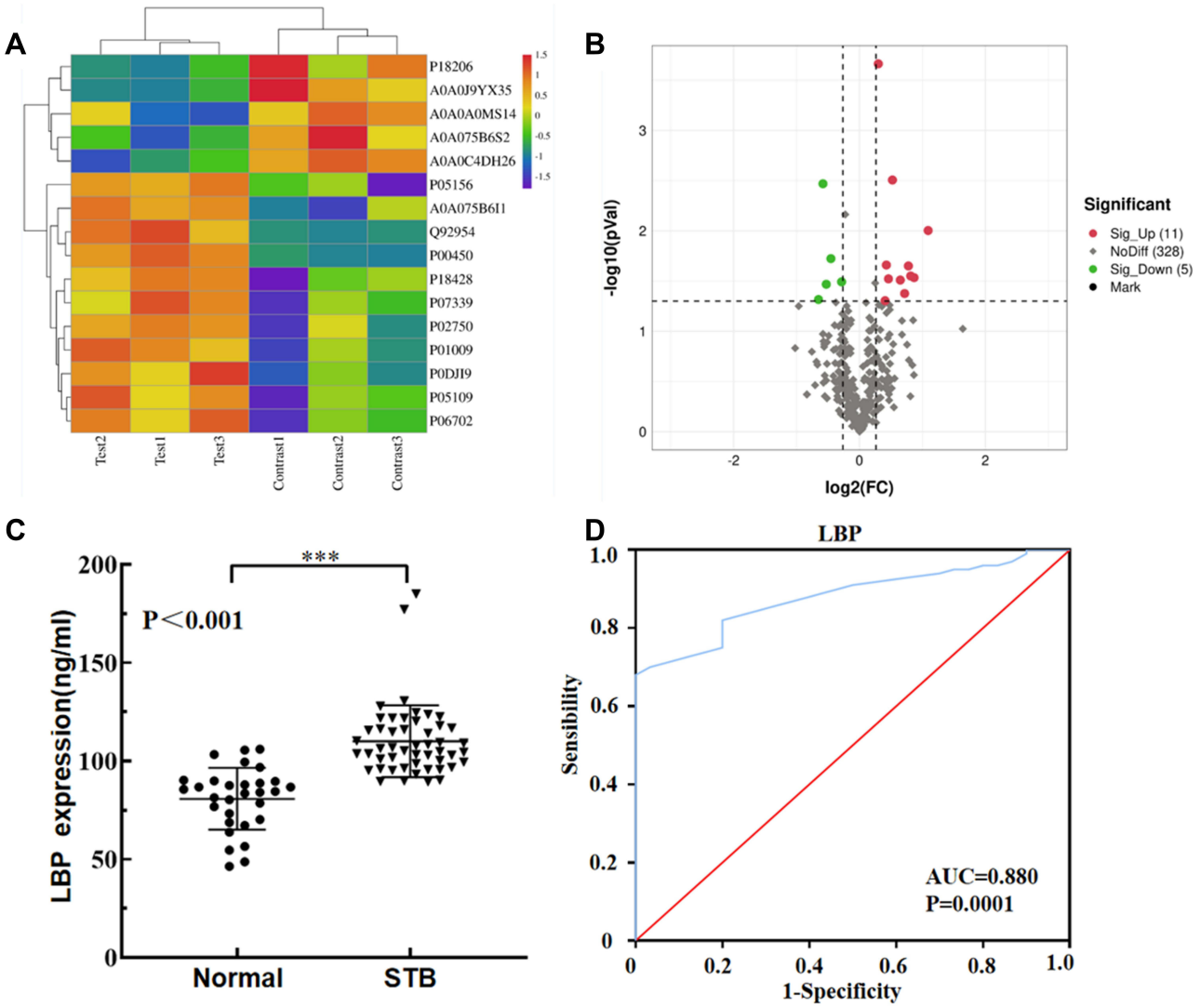

Figure 1 Validation of LBP protein screening in peripheral blood of normal human and STB patients.
Notes: (**A**) Differential protein clustering analysis graph. (**B**) Differential protein volcano distribution. (**C**) ELISA validation of differential protein LBP result analysis (***P < 0.001, the difference was statistically significant; Normal: healthy people). (**D**) ROC curve results of LBP in STB examination.
Abbreviations: FC, fold change; STB, spinal tuberculosis; LBP, lipopolysaccharide-binding protein; AUC, area under the curve.

pathway and Apoptosis may play important roles in the development of STB. These results indicated that multiple functions and signaling pathways of cells participate in the occurrence or development of STB. Meanwhile, significantly different proteins from the screening were associated with TLRs and NF-κB signaling pathways (Figure 2A and B).

Results of Laboratory Tests

In the experimental examination index, the value measurements of healthy people and STB patients were compared and found that there was no statistical difference in age, WBC, NEUT%, LYM% and other aspects. However, the ESR and CRP measurements in STB patients were significantly higher than those in normal patients. As we all know, ESR and CRP are important test indicators for evaluating STB patients. The LBP measurements (102.73±23.29) in STB patients were significantly higher than the normal LBP measurements (81.30±15.88), and there were statistically significant differences between the two groups, so LBP may be used as an indicator to evaluate the difference between STB patients and normal people (Table 3).

Table 1 Results of GO Functional Annotation Analyses

GO Term	Description	Input Number	*P* value
GO:0035662	Extracellular space	16	0.001
GO:0044548	Localization	5	0.001
GO:0050544	Response to stimulus	14	0.001
GO:0050786	Biological Regulation	13	0.001
GO:0006805	Cellular process	13	0.001
GO:0006914	Locomotion	16	0.001
GO:0006919	Multi-organism process	15	0.001
GO:0032119	Multicellular organismal process	9	0.001
GO:0044057	Developmental process	9	0.001
GO:0051000	Growth	5	0.001
GO:0051493	Metabolic process	6	0.001
GO:0050729	Cell killing	1	0.001
GO:0001726	Signaling	6	0.001
GO:0016529	Cellular component organization or biogenesis	11	0.001
GO:0030017	Reproductive process	11	0.001
GO:0008017	Reproduction	10	0.001
GO:0002224	Leukocyte migration	13	0.001
GO:0030307	Cell growth	15	0.001
GO:0050832	Immune response	8	0.001

Table 2 Results of KEGG Signaling Pathway Analyses

Protein ID	Definition	*P* value	Signaling Pathway
P18428	Lipopolysaccharide-binding protein	0.001	NF-kappaB Toll-like receptor
P05109	Protein S100-A8	0.015	IL-17
P06702	Protein S100-A9	0.044	IL-17
P05156	Complement factor I	0.023	NF-kappaB
P01009	Alpha-1-antitrypsin	0.015	Complement and coagulation cascades
P07339	Cathepsin D	0.072	Tuberculosis
P00450	Ceruloplasmin	0.035	Porphyrin and chlorophyll metabolism
P18206	Vinculin	0.001	Regulation of actin cytoskeleton
A0A0J9YX35	Immunoglobulin heavy chain	0.026	PI3K-Akt
A0A0A0MS14	Immunoglobulin heavy chain	0.009	Calcium

Analysis of LBP in Clinical Diagnosis

Differential expression in serum of STB patients was measured by ELISA method, and the results of patients in age, sex, course, WBC, NEUT%, LYM%, pathology, T-SPOT, acid fast bacillus and imaging were analyzed, and the results showed that STB patients had no statistical significance ($P > 0.05$) in age, sex, course, WBC, NEUT, LYM%, and ESR, CRP, pathology, T-SPOT. There were statistically significant results for acid-resistant staining and imaging ($t \leq 0.05$) (Table 4).

Analysis of LBP Protein Measurement Data in Laboratory Examination

Correlation analysis of clinical counts was carried out using curve regression and Spearman correlation analysis, and it was found that the LBP protein content in patients was positively correlated with CRP and ESR changes, which was statistically significant ($P \leq 0.01$), and there was no correlation with the course, age, WBC, NEUT%, LYM% values, and there was no statistical significance ($P \geq 0.05$) (Figure 3 and Table 5). It is suggested that LBP has a synergistic effect with CRP and ESR in the detection of infectious or inflammatory diseases.

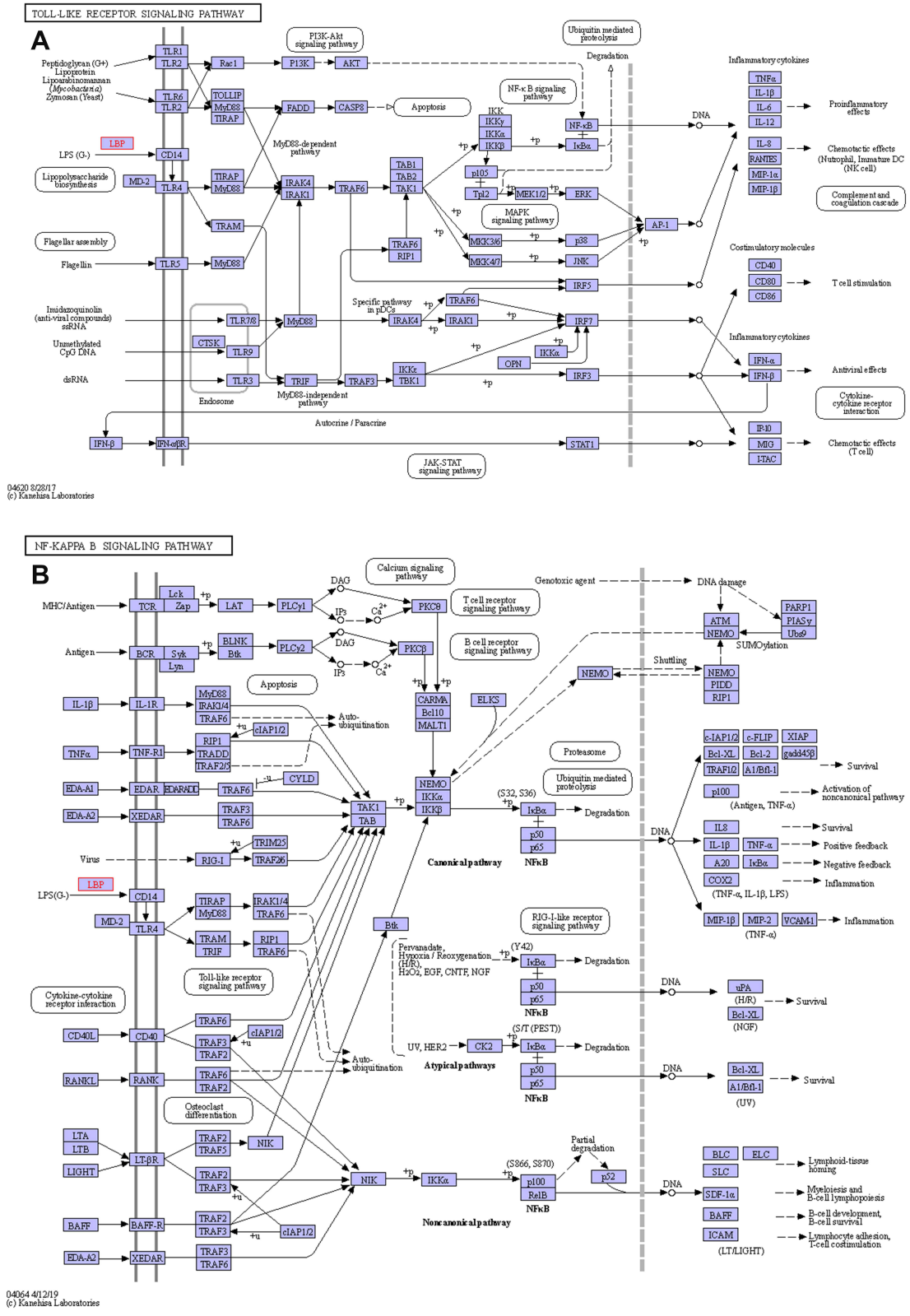

Figure 2 Screening of LBP-related signalling pathways by KEEGG analysis.
Note: (**A**) LBP and TLRS-related signaling pathway, (**B**) LBP and NFκB-related signaling pathway.

Table 3 General Statistics of Healthy Individuals and STB Patients

Item (x±s)	Healthy Person (n=30)	STB Patients (n=100)
Age (years)	53.39±9.67	49.47±16.33
Course (days)	NA	28.88±16.07
LBP (ng/mL)	81.30±15.88	102.73±23.29
CRP (mg/L)	1.11±0.70	24.92±23.69
ESR (mm/h)	8.27±4.20	40.48±28.71
WBC (*10^9/L)	5.99±1.28	6.61±1.84
NEUT%	60.19±5.02	64.72±10.93
LYM%	23.65±7.86	24.13±9.53

Abbreviations: CRP, C-reactive protein; ESR, erythrocyte sedimentation rate; WBC, white blood cell; NEUT, neutrophilic granulocyte; LYM, lymphocyte.

Table 4 Distribution of LBP Proteins in STB Clinical Phenotypes

Item	n	LBP Value (x±s)	*t*-Test (*t*)	*P*
Age (years)				
≤50	51	103.27±20.54	0.237	0.813
>50	49	102.16±26.05		
Gender				
Men	50	101.15±23.74	−0.678	0.499
Women	50	104.31±22.96		
Course (days)				
≤14	23	100.81±32.89	−0.450	0.654
>14	77	103.30±19.80		
WBC values				
Normal	91	101.66±22.61	−1.475	0.143
Abnormal	9	113.59±28.58		
LYM %				
Normal	67	99.83±22.11	−1.792	0.076
Abnormal	33	108.61±24.82		
NEUT %				
Normal	78	100.53±22.17	−1.801	0.075
Abnormal	22	110.54±25.96		
ESR Values				
Normal	22	83.13±24.26		
Abnormal	78	108.26±19.90	−4.978	0.000
CRP Values				
Normal	15	66.32±14.45		
Abnormal	85	109.15±18.06	−8.696	0.000

Abbreviations: WBC, white blood cell; CRP, C-reactive protein; LYM, lymphocyte; ESR, erythrocyte sedimentation rate; NEUT%, neutrophilic granulocyte.

Evaluation of LBP as a Diagnostic Biomarker

The positive results of pathology, bacteriological culture, T-SPOT detection, imaging diagnosis, and acid fast bacillus were statistically analyzed by independent sample *t*-test and ROC curve analysis, and the results showed that the expression of LBP protein content was statistically significant in pathology positive, bacteriological positive, T-SPOT positive, imaging diagnosis, and positive acid-resistance staining ($P < 0.01$) (Table 4). The results of ROC curve analysis showed that the AUC of the LBP protein identified ASB positive for STB in pathology, bacteriology, T-SPOT, imaging, and acid fast bacillus was 0.677, 0.707, 0.751, 0.714, and 0.656 ($P < 0.01$) (Figure 4). The results showed that the effect

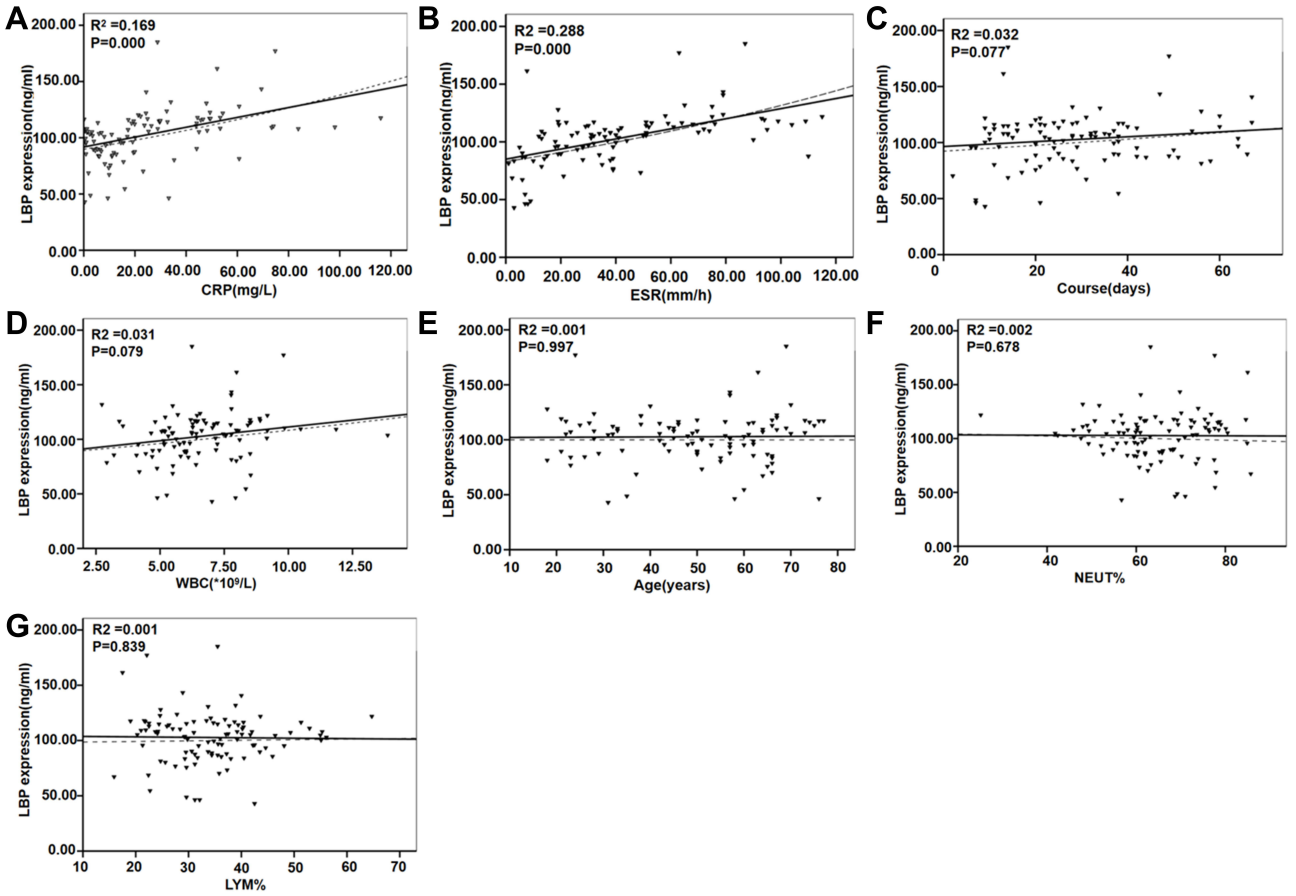

Figure 3 Analysis and detection results of LBP in peripheral blood of STB patients.
Note: (**A**–**G**) are curve regression analyses of CRP, ESR, disease duration, WBC, age, NEUT%, and LYM% values correlated with measured data of LBP expression levels in STB patients.

of LBP in diagnosing STB was related to the diagnostic results of pathology, bacteriology, T-SPOT, imaging, and acid fast bacillus, and LBP had the potential to be used as a biomarker for diagnosing STB in peripheral blood.

Discussion

STB is a common form of extrapulmonary tuberculosis, caused by Mycobacterium tuberculosis spreading through the circulatory system and localizing to the blood vessels of the bone, resulting in destructive bone and joint disease.[9] Early symptoms and clinical manifestations are atypical, and the hidden course of the disease is the main reason for the untimely treatment of STB and the main cause of serious complications, therefore diagnosis is of great significance for STB treatment and prognosis. The lack of specific and sensitive diagnostic indicators in the process of STB diagnosis and treatment is the main reason for the delay in the diagnosis and treatment of STB patients. LBP has stable and specific

Table 5 Analysis of Intra-Serum LBP Protein Levels and Clinical Phenotype in STB Patients

Spearman Rho		CRP	ESR	Course	WBC	Age	NEUT%	LYM%
LBP	Correlation coefficient	0.544	0.345	0.101	0.260	0.037	0.035	−0.064
	P-value	0.000	0.000	0.317	0.009	0.713	0.732	0.530
	N	100	100	100	100	100	100	100

Note: $P < 0.01$, significant correlation.
Abbreviations: CRP, C-reactive protein; ESR, erythrocyte sedimentation rate; WBC, white blood cell; NEUT, neutrophilic granulocyte; LYM, lymphocyte.

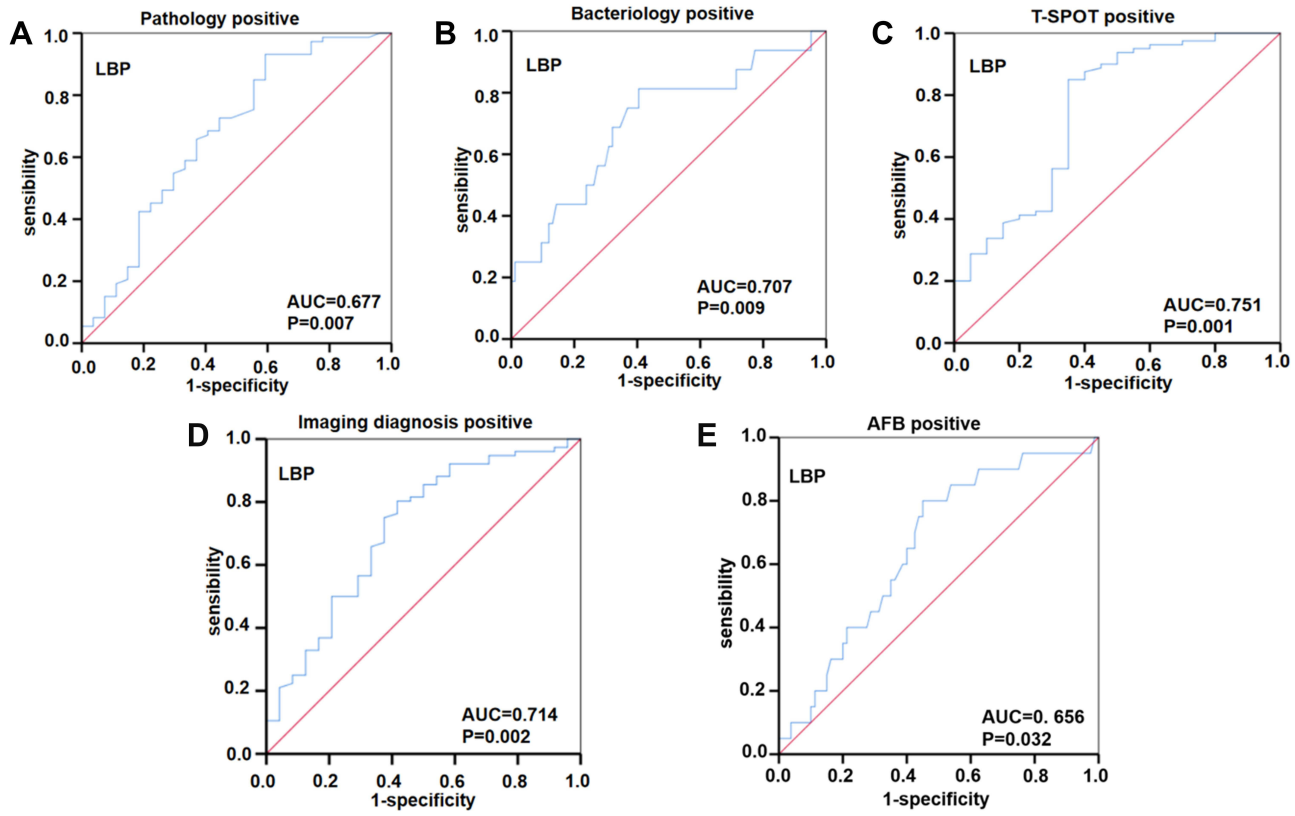

Figure 4 ROC analysis of molecular biomarkers of LBP in peripheral blood of STB patients.
Note: (**A**–**E**) are the ROC curve analysis of LBP combined with pathology, bacterial culture, T-SPOT, imaging, and antacid staining for diagnosis of STB.

expression characteristics in the peripheral blood of STB patients, which makes LBP may have special significance in STB diagnosis and potential as a diagnostic marker of STB.

In this study, we used proteomics to screen for differentially expressed proteins in the peripheral blood of STB patients and healthy individuals, under the conditions of fold change (FC) ≥ 1.5, $P<0.01$, and relevant proteins associated with STB signaling pathway, we verified by ELISA whether there was differential expression in the peripheral blood of healthy individuals and STB patients and finally determined that LBP met the conditions. Studies have shown that LBP is a protein with high affinity for binding to the acute phase of glycosylation of the bacterial lipopolysaccharide part and plays an important role in the pathophysiological process of sepsis.[10] At the same time, Gyula found that LBP expression in postoperative renal cell carcinoma patients has important implications for the detection and adjuvant treatment of the disease and can be used as a relevant biomarker for the progression of renal cell carcinoma after surgery.[6] In addition, Huang found that LBP in osteoarthritis of the knee can be used as a diagnostic marker for evaluating the severity of osteoarthritis of the knee.[11] Increased LBP levels during inflammation, insulin resistance, and metabolic stress suggest impaired cellular differentiation.[12] Elevated LBP levels have a special significance in clinical disorders, such as LBP can be used as a biomarker to determine the severity of sepsis.[10] Meanwhile, we found that LBP protein was mainly associated with TLRS signaling pathway and NF-κB signaling pathway by KEGG analysis. The study found that LBP is mainly transmembrane recognition by TLRs-related receptors, which plays a role in detecting and defending against pathogenic microorganisms in immunity, and high concentrations of LBP in the host are believed to play a protective role in immune defense.[13] The classic NF-κB signaling pathway in STB pathogenesis is closely related to activation of TLRs receptors.[14] Proteomics detection and confirmation of stability differences in the expression of LBP in the peripheral blood of STB patients, therefore, it is hypothesized that LBP may provide a feedback to the organism through its increased expression in response to infection or inflammation, regulating the organism's defense function. ROC analysis showed that LBP measured the area under the curve in the peripheral blood of STB patients with an area of 0.880 (P <

0.01), confirming that changes in LBP expression in STB peripheral blood make there a close relationship or special biological function between the two. In addition, the differential expression of LBP in many clinical diseases has also been shown to be associated with the occurrence of diseases, so it is speculated that LBP has the potential to be used as a diagnostic biomarker when detecting STB. This study found differential expression in peripheral blood between LBP and STB patients, and the relationship between LBP and STB was also reported for the first time.

After determining the value of LBP in the diagnosis of STB, we further analyzed its relevance to the clinical features of STB and found that the relative expression of LBP as a detection index in peripheral blood was positively correlated with ESR and CRP levels ($P < 0.01$). It is comparable to Wen,[15] who reported that LBP is significantly correlated with elevated ESR and CRP in active disease, and has comparable value for disease prediction with ESR and CRP. There was no correlation between LBP and age, course, WBC, NEUT%, and LYM% ($P>0.05$), which was basically consistent with the evaluation index of clinical STB. LBP performed a curve regression analysis of the positive results of pathology, bacteriological culture, T-SPOT, imaging and acid fast bacillus confirmed by STB, and found that the AUC between the two was 0.677, 0.707, 0.751, 0.714 and 0.656 ($P < 0.01$), indicating that LBP is valuable for STB diagnosis. Positive pathology and cell culture are the gold standards for the definitive diagnosis of STB, and ROC curve analysis was performed on STB patients with pathology and bacteriology positive, which found that LBP had a good correlation with pathology and bacteriological culture, confirming that LBP can be used for the diagnostic evaluation of STB. T-SPOT has a significant correlation of 88% and 90% in the sensitivity and specificity of diagnosing spinal tuberculosis,[16] and LBP has an area of 0.751 under the T-SPOT curve, indicating that LBP has a significant correlation with T-SPOT, which has good sensitivity and specificity for detecting STB and can be used as a diagnostic marker for the diagnosis of STB. MRI and CT in imaging have higher sensitivity and specificity in the diagnosis of STB,[17] and there is also a good correlation in the analysis of the ROC curve of LBP in the diagnosis of MRI and CT, which indicates that LBP has high diagnostic value for STB and can be used as an evaluation factor for its diagnosis. The specificity and sensitivity of AFB in the detection of STB were 87.0% and 85.9%, respectively, and the positive curve regression results of LBP showed that the two were correlated, which had a synergistic effect in the diagnostic evaluation of STB. Combined with the reports that LBP as a biomarker for evaluating rheumatoid arthritis sensitivity, evaluating the prediction of cardiac insufficiency prognosis after radiation therapy for breast cancer, and distinguishing mastocytosis, there are significant differences in peripheral blood expression of LBP in many clinical diseases, which indicates that LBP is indeed closely related to clinical diseases.[15,18,19] In this study, the statistical analysis of the differences and clinical phenotypes of LBP in the peripheral blood of STB patients was screened and found that it has the potential as a biomarker in the diagnosis of STB.

Conclusion

In conclusion, this study found for the first time that LBP has stable differential expression in the peripheral blood of STB patients, which has high diagnostic value in the analysis of ESR, CRP, pathology and imaging indicators, and has the potential as a molecular marker for STB diagnosis; LBP may also play an important role in the study of STB pathogenesis-related mechanisms, but the specific functional mechanism still needs to be further explored. The shortcoming of this study is that the sample size collected is relatively small, LBP protein needs to be validated as a diagnostic biomarker for clinical use in STB with expanded samples.

Abbreviations

LBP, lipopolysaccharide-binding protein; STB, Spinal tuberculosis; WBC, white blood cell; CRP, C-reactive protein; ESR, Erythrocyte Sedimentation Rate; T-SPOT, T-cell spot test for tuberculosis infection; ELISA, enzyme-linked immunosorbent assay; AUC, area under the curve; TMT, Tandem Mass Tag; AFB, acid fast bacillus; MRI, Magnetic Resonance Diffusion Tension Imaging; CT, computerized tomography; NEUT, neutrophilic granulocyte; LYM, lymphocyte.

Data Sharing Statement

The datasets used and analyzed during the current study are available from the corresponding author upon reasonable request.

Ethics Approval and Consent to Participate

Approval for the study was obtained from the ethics committee of General Hospital of Ningxia Medical University. The verbal consent was obtained from each patient before enrollment. In an outbreak investigation situation, the ethics committee approved this procedure to conduct an investigation after obtaining verbal consent. All procedures were performed in accordance with relevant guidelines in the manuscript.

Author Contributions

Caili Lou is the first author. All authors made a significant contribution to the work reported, whether that is in the conception, study design, execution, acquisition of data, analysis and interpretation, or in all these areas; took part in drafting, revising or critically reviewing the article; gave final approval of the version to be published; have agreed on the journal to which the article has been submitted; and agree to be accountable for all aspects of the work.

Funding

National Natural Science Foundation of China (grant No. 81860395); Key R&D program of the autonomous region (grant No.: 2022BEG03099); Natural Science Foundation of Ningxia (No. 2020AAC03391); The National Natural Science Foundations of China (No.82160305); The First-Class Discipline Construction Project of Ningxia Medical University Clinical Medicine (2022); The fifth group of Ningxia Young Scientific and Technological Talents lifting Project (NXKJTJGC2020080).

Disclosure

The authors declare that they have no competing interests.

References

1. Chakaya J, Khan M, Ntoumi F, et al. Global tuberculosis report 2020 - reflections on the global TB burden, treatment and prevention efforts. *Int J Infect Dis*. 2021;1:S7–S12. doi:10.1016/j.ijid.2021.02.107
2. Yang L, Hao D, Yunxuan L, et al. Analysis of risk factors associated with surgical site infection after spinal tuberculosis. *Chin J Spinal Cord*. 2020;11:964–969.
3. Qiao B, Chen Z, Huang J, et al. Lipopolysaccharide-binding protein as a biomarker in oral and maxillofacial tumors. *Oral Dis*. 2021;321:11–17.
4. Kopp F, Kupsch S, Schromm AB. Lipopolysaccharide-binding protein is bound and internalized by host cells and colocalizes with lps in the cytoplasm: implications for a role of lbp in intracellular lps-signaling. *Biochim Biophys Acta*. 2016;4:660–672. doi:10.1016/j.bbamcr.2016.01.015
5. Asada M, Oishi E, Sakata S, et al. Serum lipopolysaccharide-binding protein levels and the incidence of cardiovascular disease in a general Japanese population: the hisayama study. *J Am Heart Assoc*. 2019;8:13628. doi:10.1161/JAHA.119.013628
6. Kovacs G, Peterfi L, Farkas N, et al. Expression of inflammatory Lipopolysaccharide Binding Protein (Lbp) predicts the progression of conventional renal cell carcinoma - a short report. *Cell Oncol*. 2017;6:651–656. doi:10.1007/s13402-017-0346-4
7. Boylan KL, Andersen JD, Anderson LB, et al. Quantitative proteomic analysis by Itraq (R) for the identification of candidate biomarkers in ovarian cancer serum. *Proteome Sci*. 2010;8:31. doi:10.1186/1477-5956-8-31
8. Sarvdeep Singh D, Vishal K, Sarvdeep Singh D, Vishal K. *Tuberculosis of the Spine[M]*. Singapore: Springer Nature; 2022.
9. Dexin H, Min W, Lijuan Z, et al. Relationship between Ifn- γ gene polymorphism and spinal tuberculosis susceptibility and serum IL-4 and Ifn- γ. *Chin J Nosocomiol*. 2020;04:568–571.
10. Aldag J, Persson T, Hartmann RK. 2'-fluoro-pyrimidine-modified RNA aptamers specific for Lipopolysaccharide Binding Protein (Lbp). *Int J Mol Sci*. 2018;12:3883.
11. Huang ZY, Perry E, Huebner JL, et al. Biomarkers of inflammation lbp and tlr predict progression of knee osteoarthritis in the doxy clinical trial. *Osteoarthr Cartil*. 2018;12:1658–1665. doi:10.1016/j.joca.2018.08.005
12. Moreno-Navarrete JM, Jove M, Padro T, et al. Adipocyte Lipopolysaccharide Binding Protein (Lbp) is linked to a specific lipidomic signature. *Obesity*. 2017;2:391–400. doi:10.1002/oby.21711
13. Fish-Low CY, Than L, Ling KH, et al. Plasma proteome profiling reveals differentially expressed lipopolysaccharide-binding protein among leptospirosis patients. *J Microbiol Immunol Infect*. 2020;1:157–162. doi:10.1016/j.jmii.2018.12.015
14. Brennan JJ, Gilmore TD. Evolutionary origins of toll-like receptor signaling. *Mol Biol Evol*. 2018;7:1576–1587. doi:10.1093/molbev/msy050
15. Wen W, Li Y, Cheng Y, et al. Lipopolysaccharide-binding protein is a sensitive disease activity biomarker for rheumatoid arthritis. *Clin Exp Rheumatol*. 2018;2:233–240.
16. Yao L, Jian S, Bing W, et al. A meta-analysis of the diagnostic value of T cell spot test for peripheral blood tuberculosis infection in spinal tuberculosis. *Chin J Tuberculosis*. 2019;10:1084–1089.
17. Chun C, Yong Z, Chao Y, et al. Magnetic resonance imaging; 2016;5:371–375.
18. Chalubinska-Fendler J, Graczyk L, Piotrowski G, et al. Lipopolysaccharide-binding protein is an early biomarker of cardiac function after radiation therapy for breast cancer. *Int J Radiat Oncol Biol Phys*. 2019;5:1074–1083. doi:10.1016/j.ijrobp.2019.04.002
19. Graham R, McMullen AA, Moore G, et al. Swath-Ms identification of Cxcl7, Lbp, tgfbeta1 and pdgfrbeta as novel biomarkers in human systemic mastocytosis. *Sci Rep*. 2022;1:5087. doi:10.1038/s41598-022-08345-3