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THE INFLUENCE OF ADVANCED GLYCATION END PRODUCTS (AGES) IN PACIENTS WITH PULMONARY TUBERCULOSIS AND DIABETES MELLITUS

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INTRODUCTION. *The coexistence of diabetes mellitus (DM) and pulmonary tuberculosis (TB) represents a major global public health challenge, with diabetes being recognized as an important risk factor for the development and progression of TB. Chronic hyperglycemia promotes the formation and accumulation of advanced glycation end products (AGE), which contribute to oxidative stress, persistent inflammation, and immune dysfunction. In patients with TB-DM comorbidity, elevated AGE levels may impair innate and adaptive immune responses by reducing phagocytic activity, altering macrophage function, and delaying antigen presentation, thereby favoring the persistence and severity of Mycobacterium tuberculosis infection.*

OBJECTIVE. *To evaluate the influence of advanced glycation end products (AGE) in patients with pulmonary tuberculosis and diabetes mellitus and to determine their role in the inflammatory and immune response associated with TB-DM comorbidity.*

MATERIAL AND METHODS. *The prospective observational study was conducted at the "Chiril Draganiuc" Institute of Pneumology during the period 2019–2024 and included 118 patients. The study population was divided into two subgroups: group L_1, consisting of 59 patients with pulmonary tuberculosis and diabetes mellitus, and group L_2, comprising 59 patients with pulmonary tuberculosis without diabetes mellitus. All participants were assessed before and after in-hospital treatment. The diagnosis of pulmonary tuberculosis was confirmed by acid-fast bacilli (AFB) smear microscopy and the molecular-genetic method Xpert MTB/RIF. In order to evaluate the influence of advanced glycation end products (AGEs) in TB-DM comorbidity, serum AGE levels, biomarkers of systemic inflammation like Ceruloplasmin and C-reactive protein were determined and comparatively analyzed between the study groups.*

RESULTS. *In the study, we demonstrated importance of AGEs and systemic inflammatory markers Ceruloplasmin and C-reactive protein (CRP) showed distinct dynamics during treatment. In the TB-DM group, ceruloplasmin exhibited a more moderate increase of 19.3%, compared to 30.8% in group 1. Diabetes mellitus was also associated with a lower overall rise in serum AGE levels, reaching 153% by the end of therapy. Regarding bacteriological outcomes, the proportion of patients with negative AFB results decreased 2.3-fold in the study group during treatment, whereas in group 1 the reduction was more pronounced, reaching 5.4-fold, with a decline from 62.7% to 27.1%. A similar trend was observed for Xpert MTB/RIF, with a decrease in the number of negative patients in both groups throughout therapy.*

Overall, the coexistence of these two conditions is characterized by a more pronounced attenuation of the immune–inflammatory response during anti-tuberculosis treatment.

CONCLUSION. *Overall, these findings indicate that AGEs, ceruloplasmin, and C-reactive protein collectively represent valuable biomarkers reflecting the complex interaction between oxidative stress, inflammation, and immune dysfunction in pulmonary tuberculosis associated with type II diabetes mellitus, and may serve as useful indicators for disease severity, progression, and therapeutic response.*

Key words: ***AGEs****, Ceruloplasmin, C reactive protein, diabetes mellitus, pulmonary tuberculosis, Xpert MBT/Rif.*

INTRODUCTION

The interaction between tuberculosis and diabetes mellitus has become increasingly recognized as a significant clinical and epidemiological problem. Diabetes mellitus contributes to impaired immune defense mechanisms, thereby increasing susceptibility to Mycobacterium tuberculosis infection and favoring disease progression. Epidemiological evidence indicates that individuals with diabetes present an approximately threefold higher risk of developing active tuberculosis compared to non-diabetic populations [1,2]. Furthermore, the prevalence of diabetes among patients diagnosed with pulmonary tuberculosis varies considerably across different populations, ranging between 1.9% and 35% [3]. Conversely, tuberculosis itself may induce transient disturbances in glucose metabolism and stress-related hyperglycemia, complicating glycemic control and the management of diabetic patients [4].

According to the 2025 Global Tuberculosis Report published by the World Health Organization, approximately 8.3 million new cases of tuberculosis were reported worldwide in 2024. More than half of these patients (54%) underwent initial evaluation using rapid molecular diagnostic methods, reflecting a notable increase compared to 48% reported in 2023. Despite advances in diagnostic strategies and treatment approaches, pulmonary tuberculosis remains a major global health concern, influenced by several important risk factors, including malnutrition, HIV infection, tobacco smoking, excessive alcohol consumption, and diabetes mellitus [5]. Patients with TB-DM comorbidity frequently present with more severe clinical manifestations and unfavorable disease evolution. Several studies have demonstrated that these patients exhibit higher rates of sputum smear positivity at diagnosis, more extensive pulmonary lesions, and increased frequency of symptoms such as persistent fever, productive cough, and hemoptysis [6]. In addition, diabetes mellitus has been associated with delayed sputum conversion and prolonged persistence of positive smear microscopy and culture results during the intensive phase of anti-tuberculosis therapy, suggesting a slower therapeutic response and increased risk of adverse outcomes [7].

Chronic inflammatory diseases such as tuberculosis (TB) and diabetes mellitus (DM) are pathogenetically associated with oxidative stress. In diabetes mellitus, intrinsic predisposition factors, including aging and

family history, together with extrinsic factors such as smoking, further contribute to increased oxidative stress [8,9]. Oxidative stress in diabetic patients is closely related to the formation of advanced glycation end products (AGEs), which result from non-enzymatic chemical reactions between proteins and sugars [10]. Excessive accumulation of AGEs in patients with DM is involved in the pathophysiology of chronic diabetic complications, including diabetic cardiomyopathy through AGE-induced endothelial progenitor cell dysfunction. AGEs may exert their biological effects through both receptor-dependent and receptor-independent mechanisms. The receptor for advanced glycation end products (RAGE), a member of the major histocompatibility complex (MHC) class III family, is expressed on the surface of immune cells and plays an important role in the regulation of tuberculosis infection. In pulmonary tuberculosis, an effective immune response against Mycobacterium tuberculosis largely depends on the proper function of macrophages and T lymphocytes. AGEs interfere with these immune mechanisms by altering the structure and function of proteins and receptors involved in antigen recognition and phagocytosis. Consequently, macrophages exposed to elevated AGE levels exhibit a reduced capacity to phagocytose and eliminate Mycobacterium tuberculosis, thereby promoting persistence of the infection and progression of the disease [11].

In patients with tuberculosis–diabetes mellitus (TB-DM) comorbidity, elevated levels of advanced glycation end products (AGEs) may contribute to a more severe disease course and a delayed therapeutic response. Individuals with diabetes frequently exhibit poor glycemic control, which promotes the continuous formation and accumulation of AGEs and sustains a persistent pro-inflammatory environment. This may partially explain the increased susceptibility of these patients to extensive pulmonary lesions, complications, and recurrent tuberculosis.

From a clinical perspective, AGEs may be considered indirect biomarkers of metabolic imbalance and chronic inflammation. Although their assessment is not yet widely implemented in routine clinical practice, the inclusion of AGE evaluation in studies investigating the interaction between tuberculosis and diabetes mellitus may provide valuable insights into the underlying pathogenic mechanisms involved in TB-DM comorbidity.

An important aspect is that the reduction of AGE formation can be achieved through adequate glycemic control and therapeutic interventions targeting oxidative stress and inflammation. Therefore, optimal management of diabetes mellitus in patients with pulmonary tuberculosis may not only improve metabolic control but also enhance immune response and increase the effectiveness of anti-tuberculosis treatment [12].

MATERIALS AND METHODS

The prospective observational study was conducted at the Chiril Draganiuc Institute of Pneumology during the period 2019–2024 and included 118 patients. The study group was divided into two subgroups: group L_1, consisting of 59 patients with pulmonary tuberculosis and diabetes mellitus, and group L_2, comprising 59 patients with pulmonary tuberculosis without diabetes mellitus. All participants were assessed both before and after the in-hospital treatment. Diagnosis was based on acid-fast bacilli (AFB) smear microscopy, the molecular-genetic Xpert MTB/RIF method, AGEs indices, Ceruloplasmin and C-reactive protein levels. All patients were treated according to the national clinical protocol and were evaluated at one-month intervals during hospitalization.

The study was approved by decision no. 31 of 18.05.2019, issued by the Research Ethics Committee of Nicolae Testemițanu State University of Medicine and Pharmacy. The representative sample size was calculated using EpiInfo version 7.2.2.6, in the "Stat Calc – Sample Size and Power" module. The mean, median, and standard deviation (SD) were determined. To compare the two groups, the Student's t-test was applied, and the statistical significance of differences was assessed using the p-value, with $p < 0.05$ considered significant.

RESULTS

According to the demographic distribution, the analyzed study samples demonstrated a predominantly male distribution in both groups, more pronounced in the non-diabetes mellitus group. Female participants were underrepresented compared to males in both the diabetes mellitus and non-diabetes mellitus groups (figure1).

To more accurately determine the impact of diabetes mellitus on the clinical course of pulmonary tuberculosis and on the effectiveness of anti-tuberculosis therapy, it is important to analyze biomarkers that demonstrate significant differences between patients with TB-DM comorbidity and those without diabetes, both before and after treatment.

In order to evaluate the particularities of oxidative stress and inflammatory response in patients with pulmonary tuberculosis associated with diabetes mellitus, circulating levels of advanced glycation end products (AGEs), ceruloplasmin, and C-reactive protein (CRP) were assessed in group 1 (TB-DM) at the initiation and completion of anti-tuberculosis treatment (Table1).

Figure 1. Distribution of Pacients DZ/non DZ

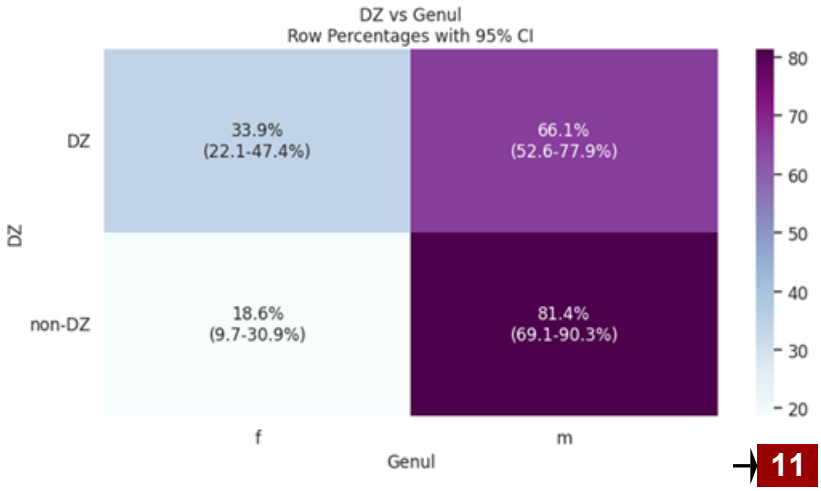

Table 1. Changes in serum levels (M±SD) of AGEs and inflammatory markers

Marker	Pacients TB-DM study group (n=59)		± (%)	p
	Before treamtent	After treatment		
AGEs (pentosidine), µmol/L Median	5996,4±889 2119	15208±12622 25979	+153%	<0.00031
Ceruloplasmin, µg/L Median	828,6±256.6 832,3	988,7±288.4 1003.8	+19,3%	<0.00286
PCR, mg/L Median	32,1±43 12	23,3±51 12	-47,8%	0,1172

Note: *p – significance value comparing after treatment vs. before treatment; ± (%) – relative changes of the marker during the treatment period.*

Table 2. Changes in serum levels (M±SD) of AGEs and inflammatory markers in patients with TB and DM over a one-month treatment period

Marker	Pacients TB control group (n=59)		± (%)	p
	Before treamtent	After treatment		
AGEs (pentosidine), µmol/L Median	2030,3±508,4 2237,9	26684,3±526,8 26732	elevated>13 times	<0.000
Ceruloplasmin, µg/L Median	687,8±233 727,7	899,2±220 881,2	+30,85%	<0.000
PCR, mg/L Median	27,4±45,9 12	14,3±13,9 14,7	-47,8%	0,006

Note: *p – significance value comparing after treatment vs. before treatment; ± (%) – relative changes of the marker during the treatment period*

Table 3. Median values of AFB smear and Xpert MTB/RIF indices before and after a one-month treatment period

Indice	Before treatment				After treatment			
	Grup$_1$ DZ vs group$_2$ non DZ				Grup$_1$ DZ vs group$_2$ non DZ			
	DZ n (%)	-DZ N (%)	DZ vs -DZ	p	DZ n (%)	-DZ n (%)	DZ vs -DZ	P
AFB pozitiv	37 (62,7%)	27 (45,8%)	+37%	>0,05	16 (27,1%)	5 (8,5%)	+220%	0,014
Xpert MTB/ Rif negativ	10 (16,9%)	23 (39%)	-56,5%	0,013	26 (40,7%)	37 (62,7%)	-29,8%	0,026

Notably, the reduction in serum CRP levels was similar in both groups: 47.8%. In contrast to the positive dynamics of Ceruloplasmin in group$_1$, was a more moderate, yet significant, elevation in ceruloplasmin levels, with an estimated increase rate of 19.3% (compared to 30.8% in group 1). In addition, the association with diabetes mellitus was characterized by a considerably more limited increase in serum AGE levels; by the end of treatment, the rise of this marker reached 153%.

Additionally, we evaluated AGEs, Ceruloplasmin, and C-Reactive Protein Parameters in the Control Group (Table 2).

The serum level of ceruloplasmin, a carrier of bivalent copper with pro-oxidative properties, also increased significantly by 30.85%. These findings suggest an intensification of ceruloplasmin activity under the studied conditions. A combined interpretation of these two markers, C-reactive protein (CRP) and ceruloplasmin, indicates a balance between pro-inflammatory and anti-inflammatory processes: the increase in ceruloplasmin reflects activation of antioxidant and defense mechanisms, while the reduction in CRP suggests attenuation of acute inflammation. This correlation points to the organism's adaptive response to the studied conditions and highlights the usefulness of both markers in the assessment of inflammatory and oxidative status.

A remarkable increase in advanced glycation end products was also observed. Thus, pentosidine, a specific prototype of AGEs, increased in blood more than 13-fold at the end of treatment compared to the baseline level. Current pathophysiological concepts associate elevated pentosidine levels with diabetic hyperglycemia and/or oxidative stress, emphasizing a specific mechanism of somatic and immune cell stimulation mediated through the RAGE receptor. This pronounced augmentation suggests the active involvement of advanced glycation processes in the pathogenesis of pulmonary tuberculosis, being associated with intense oxidative stress and chronic inflammation characteristic of the disease. The dramatic increase in AGEs may contribute to functional impairment of pulmonary tissues through the formation of covalent cross-links between proteins, thereby affecting the elasticity and integrity of the extracellular matrix. Furthermore, AGE accumulation may amplify the local inflammatory response, creating a vicious cycle between glycation, oxidative stress, and inflammation, which supports the monitoring of this parameter as a marker of disease progression and severity.

In our study, we also evaluated the dynamics of laboratory tests, including AFB smear and Xpert MTB/RIF. To conclusively assess the impact of diabetes mellitus on the course of TB and, in particular, on the effectiveness of the applied therapy, it is informative to analyze

markers showing significant discrepancies between groups (DM vs. non-DM) both at baseline and after treatment.

Beneficial changes in AFB smear and Xpert MTB/RIF indices were observed in both groups, but their relative restoration was superior in the group without diabetes mellitus (Table 3).

The proportion of AFB-positive smears decreased by 56.8% after treatment in the DM group, while the corresponding reduction in the non-DM group was 81.2%. Thus, the initially non-significant discrepancy became significant after treatment ($p < 0.014$), and the relative incremental gap between DM and non-DM increased from 37% to 220%.

The proportion of negative Xpert MTB/RIF results increased during anti-TB treatment in both groups, indicating the effectiveness of the therapy in the context of this test's ability to detect or rule out the presence of mycobacteria in biological samples. The reduction in AFB-positive smears correlates with the decline in positive Xpert MTB/RIF results. However, although the increase in negative Xpert MTB/RIF results after treatment was higher in the DM group (160% vs. 61%), the final parameter remained significantly higher (+42.3%) in the group without diabetes mellitus.

DISCUSSION

The impact of tuberculosis (TB) on morbidity, disability, and mortality rates within the human community remains a major medical and social concern, particularly in countries with limited economic resources. In the spectrum of mortality caused by infectious diseases, TB is surpassed only by COVID-19, which ranks first according to World Health Organization data, while in 2024 the number of deaths caused by TB was estimated at 1.23 million [13,14]. Although the pulmonary system represents the primary target of Mycobacterium tuberculosis infection, other vital organs are also susceptible, including bones, the brain, and the eyes [15].

Advanced glycation end products (AGEs) and ceruloplasmin represent important biomarkers involved in the pathophysiological mechanisms of pulmonary tuberculosis associated with diabetes mellitus. Their evaluation provides valuable information regarding the intensity of oxidative stress, chronic inflammation, and metabolic imbalance that characterize TB-DM comorbidity. The combined assessment of AGEs and ceruloplasmin provides a broader understanding of the balance between oxidative damage and compensatory antioxidant mechanisms in TB-DM patients. While elevated AGE levels indicate intensified glycation processes and chronic oxidative stress, increased ceruloplasmin may represent an adaptive protective response aimed at limiting tissue injury. The coexistence of these changes highlights the complex interaction between metabolic disorders, inflammation, and immune dysfunction in pulmonary tuberculosis associated with diabetes mellitus.

From a clinical perspective, monitoring AGEs and ceruloplasmin may contribute to the evaluation of disease severity, therapeutic response, and risk of complications in patients with TB-DM comorbidity. These biomarkers may also represent potential therapeutic targets, particularly through interventions focused on improving glycemic control, reducing oxidative stress, and modulating chronic inflammation.

In our study, the impact of diabetes mellitus (DM) on the effectiveness of specific TB therapy was assessed by evaluating AFB smear and Xpert MTB/RIF values before and after treatment. It is noteworthy that in $group_2$, the number of patients with negative AFB smears decreased 2.3-fold during treatment (in $group_1$, the reduction reached 5.4-fold), with the relative decline estimated from 62.7% to 27.1%. Thus, the number of AFB-positive patients among those with TB and DM remained statistically higher compared to $group_1$. At the same time, the reduction in AFB-positive cases was associated with an increase in negative Xpert MTB/RIF results from 16.9% to 40.7%, with the final value still lower than that in $group_1$ (40.7% vs. 62.7%).

Regarding the inflammatory status, it is worth noting the higher serum CRP levels in $group_2$ (TB+DM) compared to $group_1$, both at baseline (by 17.1%) and at the end of treatment (by 62.9%). The degree of elevation of this general inflammation marker reflects the severity of the infectious agent's manifestation and serves as a negative predictor of TB progression [16]. In group2, the increased CRP synthesis, determined by the functional activity of *Mycobacterium tuberculosis*, is compounded by the pro-inflammatory effect of type II diabetes mellitus, which also involves extrapulmonary cells, including vascular endothelial cells. Similarly, the increase in ceruloplasmin, the carrier of bivalent copper with pro-oxidative properties, was lower: 19.3% versus 30.85%. The elevation of ceruloplasmin was accompanied by changes in C-reactive protein (CRP), both markers representing acute-phase proteins. Pro-oxidative activity is regulated by the antioxidant system, the functional capacity of which was assessed through the determination of serum levels of several specific biomarkers.

In this context, it is important to highlight the opinion of S. Masafumi et al. (2022), who stated that elevated CRP levels at admission—i.e., before the start of anti-TB treatment—pose a hazard for TB-related complications [17].

CONCLUSIONS

In conclusion, the association of type II diabetes mellitus with pulmonary tuberculosis is characterized by a dysregulated immune–inflammatory response during anti-tuberculosis treatment, closely linked to alterations in oxidative stress and metabolic imbalance. This is evidenced by persistently elevated levels of advanced glycation end products (AGEs), reflecting enhanced glycation processes and sustained oxidative stress, together with significant changes in ceruloplasmin and C-reactive protein (CRP), both key acute-phase proteins involved in systemic inflammatory regulation.

The observed patterns suggest an imbalance between pro-oxidant and antioxidant mechanisms, where increased AGEs contribute to immune dysfunction and impaired host defense, while variations in ceruloplasmin indicate activation of compensatory antioxidant responses.

In parallel, CRP dynamics reflect the intensity and resolution of the systemic inflammatory process.

Therapeutic efficacy, evaluated in both groups through AFB smear microscopy and Xpert MTB/RIF molecular testing, demonstrates the negative impact of diabetes mellitus on tuberculosis treatment outcomes. Patients with diabetes showed higher rates of persistent bacteriological positivity at both baseline and the end of therapy, likely associated with delayed mycobacterial clearance due to impaired immune-inflammatory responses and persistent oxidative stress. Similarly, slower normalization of CRP levels and sustained elevation of AGEs further support a prolonged inflammatory and metabolic disturbance in the TB-DM group.

References

1. Nyirenda JLZ, Wagner D, Ngwira B, Lange B. *Bidirectional screening and treatment outcomes of diabetes mellitus (DM) and Tuberculosis (TB) patients in hospitals with measures to integrate care of DM and TB and those without integration measures in Malawi.* BMC Infect Dis. 2022 Jan 4;22(1):28.
2. Quist-Therson R, Kuupiel D, Hlongwana K. *Mapping evidence on the implementation of the WHO's collaborative framework for the management of tuberculosis and diabetes: a scoping review protocol.* BMJ Open. 2020 Jan 21;10 (1): e033341.
3. Noubiap JJ, Nansseu JR, Nyaga UF, Nkeck JR, Endomba FT, Kaze AD, Agbor VN, Bigna JJ. *Global prevalence of diabetes in active tuberculosis: a systematic review and meta-analysis of data from 2·3 million patients with tuberculosis. Lancet Glob Health.* 2019 Apr;7(4): e448-e460.
4. Stubbs B, Siddiqi K, Elsey H, Siddiqi N, Ma R, Romano E, Siddiqi S, Koyanagi A. *Tuberculosis and Non-Communicable Disease Multimorbidity: An Analysis of the World Health Survey in 48 Low- and Middle-Income Countries.* Int J Environ Res Public Health. 2021 Mar 2;18(5):2439.
5. Yang H, Ruan X, Li W, Xiong J, Zheng Y. *Global, regional, and national burden of tuberculosis and attributable risk factors for 204 countries and territories, 1990-2021: a systematic analysis for the Global Burden of Diseases 2021 study*. BMC Public Health. 2024 Nov 11; 24(1): 3111.
6. Workneh MH, Bjune GA, Yimer SA. *Prevalence and associated factors of tuberculosis and diabetes mellitus comorbidity: A systematic review.* PLoS One. 2017 Apr 21;12(4): e0175925.
7. Huang LK, Wang HH, Lai YC, Chang SC. *The impact of glycemic status on radiological manifestations of pulmonary tuberculosis in diabetic patients.* PLoS One. 2017 Jun 19;12(6): e0179750.
8. Ferlita S, Yegiazaryan A, Noori N, Lal G, Nguyen T, To K, Venketaraman V. Type 2 Diabetes Mellitus and Altered Immune System Leading to Susceptibility to Pathogens, Especially *Mycobacterium tuberculosis*. J Clin Med. 2019 Dec 16;8(12):2219. doi: 10.3390/jcm8122219.
9. Tiwari BK, Pandey KB, Abidi AB, Rizvi SI. Markers of Oxidative Stress during Diabetes Mellitus. J Biomark. 2013; 2013:378790. doi: 10.1155/2013/378790.
10. Al-Bari MAA, Peake N, Eid N. Tuberculosis-diabetes comorbidities: Mechanistic insights for clinical considerations and treatment challenges. World J Diabetes. 2024 May 15;15(5):853-866. doi: 10.4239/wjd.v15.i5.853.
11. Guler R, Ozturk M, Sabeel S, Motaung B, Parihar SP, Thienemann F, Brombacher F. Targeting Molecular Inflammatory Pathways in Granuloma as Host-Directed Therapies for Tuberculosis. Front Immunol. 2021 Oct 20; 12:733853. doi: 10.3389/fimmu.2021.733853.
12. Kiran D, Podell BK, Chambers M, Basaraba RJ. Host-directed therapy targeting the Mycobacterium tuberculosis granuloma: a review. Semin Immunopathol. 2016 Mar;38(2):167-83. doi: 10.1007/s00281-015-0537-x.
13. Alame-Emane, A. K. et al. The use of GeneXpert remnants for drug resistance profiling and molecular epidemiology of tuberculosis in Libreville, Gabon. Journal of clinical microbiology, JCM. 02257–02216 (2017).
14. World Health Organization (WHO). Global Tuberculosis Report 2022. Geneva, WHO, 2022. www.who.int/publications/i/item/9789240061729.
15. Comments on the WHO Global Tuberculosis Report 2024: Progress, Challenges and Innovations for Achieving End TB Strategy by 2035. Infectious Microbes & Diseases. 2025; 7(1):1-4. DOI: 10.1097/IM9.0000000000000169.
16. Gunasekaran H, Ranganathan UD and Bethunaickan R (2025) The importance of inflammatory biomarkers in detecting and managing latent tuberculosis infection. *Front. Immunol.* 16:1538127. doi: 10.3389/fimmu.2025.1538127.
17. Amaral E, Vinhaes C, Oliveira-de-Souza D et al. The Interplay Between Systemic Inflammation, Oxidative Stress, and Tissue Remodeling in Tuberculosis. In: Antioxid Redox Signal. 2021 Jan 25;34(6):471–485. doi: 10.1089/ars.2020.8124.