History of Medicine, 2024, 10(2): 972-980
DOI: https://doi.org/10.17720/byr4mc62

Original article

THE IMPACT OF MYCOBACTERIUM ON PROTEIN PROFILES IS INVESTIGATED IN THE CONTEXT OF MALNUTRITION IN INFECTED INDIVIDUALS

***Saad un Nisa Mahesar[1], Shah Muhammad Mahesar[2], Arsalan Nizamani[3], Sohail Lund Baloch[4], Autif Hussain Mangi[5], Muhammad Ishaque Bhatti[6] & Mahvish Jabeen Channa[7]**

[1]M.Phil Scholar, Institute of Biochemistry University of Sindh Jamshoro
[2]Professor, Dean IBN-E-SINA University
[3]Assistant Professor IBN-E-SINA University Mirpurkhas
[4]Nursing Instructor Beachwood,
[5]Assistant Professor, University of Sindh Jamshoro, Pakistan
[6]Assistant Professor, Indus Medical College Tando Muhammad Khan
***Corresponding author:** Saad un Nisa Mahesar
*Institute of Bio-chemistry University of Jamshoro, Pakistan
Email: aizacute2016angel@gmail.com

Abstract

Background: Tuberculosis (TB) is a major public health problem that needs extensive research efforts to address its multiple effects on individuals.
Aims and Objectives: This study inspects the biochemical alterations such as total proteins and A/G ratio associated with mycobacterium tuberculosis. The Study has been conducted in Institute of Biochemistry University of Sindh Jamshoro in coalition with Chest Centre disease Kotri, samples were collected from male and female ward of T.B from june 2022 to July 2023.
Methods A case control study constitute a total 270 subjects, randomly selected 100
Samples of T.B patients and 100 age and gender matched healthy non T.B controls were recruited for biochemical changes carried by mycobacterium in Total proteins and A/G ratio in Tuberculosis patients. Statistical analysis was done on SPSS software.
Results: The detailed analysis on biochemical profile in patients with tuberculosis as serum total proteins level mean ± SD 6.97 $\pm$ 0.63 than in controls mean ± SD 7.26 $\pm$ 1.08, with a p-value of 0.022*, serum albumin level mean ± SD 4.91 $\pm$ 1.14 than in controls mean ± SD 4.23 $\pm$ 0.52, serum globulins levels mean ± SD 2.48 $\pm$ 0.63 than in controls mean ± SD 3.03 $\pm$ 0.71, serum A/G ratio mean ± SD 1.83 $\pm$ 0.52 than in controls mean ± SD 1.43 $\pm$ 0.25 with a p value of 0.000*, indicating results are statically significant.
Conclusion: We have observed altered biochemical profile, Total protein levels were lower in patients compared to controls, Albumin levels were significantly higher in patients than in controls. Patients had lower levels of globulin compared to control, the A/G ratio was higher in patients compared to controls, indicating significant changes in protein levels associated with the

disease severity. This study will prove to be restrained with previous works and would acknowledge potential constraints and distinct propositions for advanced investigation in future.
Key words: Pulmonary Tuberculosis, Biochemical parameters, Serum albumin, Serum globulins, A/g ratio.

Introduction

Mycobacterium, which causes TB, is an intricate condition that has a significant impact on protein profile and can suppress the host immune system of the body [1]. Examining these metabolic changes is a key component of our study motivation. Our goal is to clarify the complex pathways by which Mycobacterium affects the A/G ratio, total protein concentrations. [2]. Understanding these changes can assist improve therapy regimens and provide new opportunities for therapeutic interventions. as well as we are in search of improved diagnostic accuracy.

Pathogenesis

The pathogenesis of tuberculosis (TB) involves a complex interplay between the Mycobacterium tuberculosis (Mtb) bacterium and the host immune response. This literature review provides a summary of the current understanding of TB pathogenesis. Mtb primarily infects the lungs through inhalation of aerosolized droplets containing the bacteria [3]. Upon reaching the alveoli, Macrophages in the alveoli phagocytose MTB. Nevertheless, Mtb has developed defenses to endure and proliferate inside these macrophages, which results in the spread of infection. One of the most important factors in preventing Mtb infection is the host immunological response.[4]. Upon recognition of Mtb, macrophages release pro-inflammatory cytokines and chemokines, recruiting other immune cells to the site of infection. T cells, particularly CD4+ and CD8+ T cells, are essential for the control of Mtb infection. CD4+ T cells produce cytokines such as interferon-gamma (IFN-γ), which activate macrophages to enhance their antimicrobial activity. Additionally, host factors, such as malnutrition and diabetes, can harm the immune response and increase the risk of developing active TB disease. [5]. Infected persons often experience anemia which can lead to reduced protein levels [6]. Patients may also experience malnutrition including protein deficiency due to reduced appetite and impaired nutrient absorption. Co-morbid illnesses have also diagnosed in infected persons like, diabetes, HIV, HCV that also affect protein levels [7]. This altered protein profile can impact the hosts ability to fight infections making it harder to diagnose and treat T.B. Understanding these changes is essential for developing effective treatments and biomarkers for TB [8].

Total Proteins and A/G Ratio as Biomarkers in Tuberculosis

Total protein refers to the total amount of proteins in the blood, which includes albumin and globulins. One of the major protein parts of blood, albumins are fundamental for maintaining oncotic pressure. On the other hand, globulins are a class of proteins that are additionally partitioned into alpha, beta, and gamma globulins individually [9]. While gamma globulins are a type of immunoglobulin's (antibodies) that are significant for immune response, beta and alpha globulins are engaged with the transportation of lipids, metal particles, and chemicals in tuberculosis (TB)[10]. Many research Studies have revealed that TB infection is linked with alteration in albumins and globulins, This decrease in albumin may be due to malnutrition, inflammation, and increased protein catabolism associated with the disease globulins [11]. Additionally, TB infection can lead to significant changes in the levels of specific globulins, such as an increase in acute-phase reactants like alpha-1 antitrypsin and haptoglobin [12]. To calculate A/G ratio the value obtained by dividing the serum albumin level by the serum globulin levels. It is used as an indicator of the balance between albumin and globulins in the blood. In TB, there is decrease in albumin levels and/or the increase in specific globulins and vice versa [13]. A lower A/G ratio has been related with disease severity and poor diagnosis in various illnesses. Monitoring these parameters can provide valuable information for clinicians in controlling the TB from suspected cases [14]. Total protein levels, especially albumin levels can change in tuberculosis (TB) due to a variety of causes. Acute-phase proteins are produced as a result of the inflammatory response brought on by TB, raising globulin levels. Malnutrition and protein loss are other TB side effects that can reduce albumin production [15]. When TB is severe, liver failure might further lower albumin production. Proteins may be transferred to the site of infection and inflammation during active TB. As the illness is managed by anti-TB therapy, total protein levels may rise. It is possible to employ altered total protein levels, particularly decreased albumin levels, as both diagnostic and prognostic markers in TB. Low albumin-to-globulin ratios or significantly reduced total proteins may signify a more severe or advanced illness condition [16].

Co-morbidities

Comorbidities refer to the presence of one or more surplus medical conditions or diseases alongside a primary condition. When it comes to pulmonary tuberculosis (TB), individuals often experience comorbidities that can complicate their overall health [17].

Here are some common comorbidities associated with pulmonary tuberculosis, that is HIV/AIDS, Diabetes Mellitus, smoking, alcohol use, malnutrition, COPD (Chronic Obstructive Pulmonary Disease) kidney disease, liver disease, chronic heart diseases, mental health disorders etc [18].

Methodology

The retrospective study was accepted at institute of biochemistry university of Sindh Jamshoro, Pakistan. The study title is mycobacterium has the legacy of altering the protein profile in

infected individuals, Ethical standards are followed for current study. Every participant gives written, informed consent, and the study complies with accepted ethical standards for using human beings in research, ensuring participant rights, confidentiality, and privacy. Patients diagnosed with mycobacterium (TB) infections Samples were collected from Chest Diseases Center Kotri, from June 2022 to July 2023─ Population of study includes 270 participants, 170 infected with mycobacterium infections and 100 controls healthy individuals having no mycobacterium infections, 70 samples 0ut of 170 subjects were homogenized as we collected blood sample in yellow top gel tubes, hospital is far and hilly it causes blood samples to mix promptly. Cases and controls were complemented based on relevant demographic variables such as age, gender. Study subjects comprises male as well as female between the age group 20 to 60 years. Individuals with Mycobacterium infections having all symptoms of PTB were included in this study. Patients who experienced any co-morbid illness, diabetes, liver, kidney diseases, positive viral profile were excluded from study. Patients who participated in this study were signed written consent form.

Sample collection

Approximately, 5 ml of blood was drawn by using sterile syringe from venipuncture. The collected blood samples then transferred into chemically cleaned, and labeled EDTA gold top tubes for quantitative hematological and biochemical analysis. To separate the serum from the blood samples, centrifugation was performed at 3000 rpm for 20 minutes. For the analysis of serum total proteins and A/G ratio, a KIT method was employed, and the Quantitative analysis was done using a spectrophotometer in Pathology Laboratory. Questionnaires are specially designed for this study and were administered to the subjects. The questionnaire consists of patient's bio data, demographic characteristics and clinical analysis.

Data Analysis Procedures

The collected data were analyzed by using to comprehensive statistical analysis tool (SPSS) software and excel version 2010.

Estimation of Total proteins and A/G ratio

Serum globulin levels were calculated by subtracting the albumin concentration from the total protein. All reagents employed for these analyses were products of Diag.M (private) manufactured in France, utilizing the KIT method. To calculate the A/G ratio, follow these steps:

Total serum protein = Albumin + Globulin Globulin =Total serum protein

Total proteins – Albumin = globulin

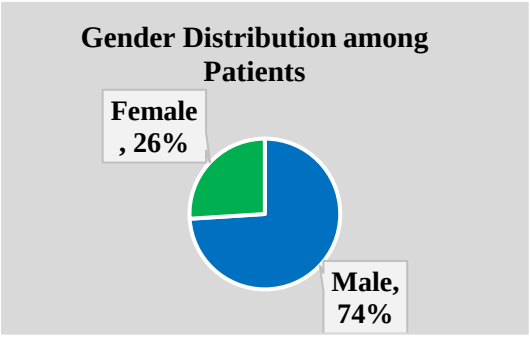
So, A/G ratio = Albumin / Globulin = / A/G ratio [19].

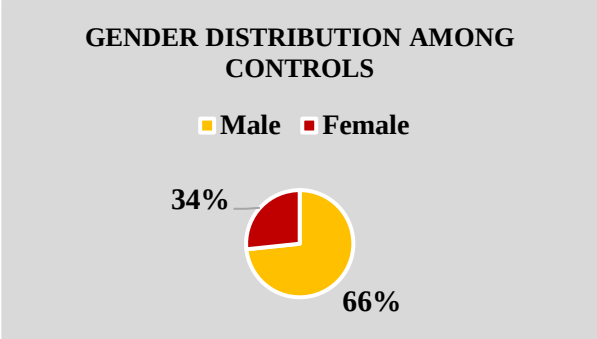
History of Medicine, 2024, 10(2): 972-980
DOI: https://doi.org/10.17720/byr4mc62

Results

By analyzing gender distribution among patients, there were **(Fig.1)** 74% male population while female comprised on 26% of population. This gender disparity highlights a notable difference in the incidence of pulmonary tuberculosis between males and females within the demographic scope of our research. In the control group, which serves as the comparative baseline for our study, there were 100 individuals without pulmonary tuberculosis. Within this group, males constituted 66% of the population with a count of 66 individuals. Females were represented by 34 individuals, accounting for 34% of the control group.

Fig.1 Gender distribution patients v/s control





CO-MORBID ILLNESS

To evaluate the comorbidities **(Table.1)** showing subset of the patient population was affected by additional health conditions alongside tuberculosis. Diabetes Mellitus was present in 10 patients, while a larger number, 21 patients, were living with Hepatitis C. A small group of 5 patients reported having both Diabetes Mellitus and Hepatitis C concurrently. However, the majority of the patients, 64 in total, did not have any of these listed comorbid illnesses. Psychological conditions were also surveyed among the TB patients. Anxiety was reported by 10 patients, whereas depression was noted in a single patient. Stress was experienced by 5 patients. Not with standing these figures, a substantial majority, 84 patients, indicated that they did not suffer from any of these psychological conditions.

TABLE NO. 1 MEDICAL HISTORY OF TB PATIENTS

VARIABLES	DESCRIPTION
COMORBID ILLNESSES	
Diabetes Mellitus	10

Hepatitis C	21
Diabetes Mellitus + Hepatitis C	5
None	64
SELF-REPORTED PSYCHOLOGICAL CONDITION	
Anxiety	10
Depression	1

The biochemical parameters of patients with pulmonary tuberculosis compared to the controls, evaluated using the t-test, for paired samples, the results indicate statistically significant differences in all measured variables, given that the p-values are all below the conventional threshold of 0.05 for significance. Total protein levels were lower in patients (mean: 6.97 g/dl) compared to controls (mean: 7.26 g/dl), with a p-value of 0.022, indicating a significant difference that could be due to the disease's impact on the nutritional status or other factors related to chronic illness. Albumin levels were significantly higher in patients (mean: 4.91 g/dl) than in controls (mean: 4.23 g/dl), with a p-value of 0.000. This is a noteworthy finding, as albumin levels are typically expected to be lower in chronic disease due to malnutrition or inflammation. Patients had lower levels of globulin (average: 2.48 g/dl) compared to people without tuberculosis (average: 3.03 g/dl), showing a big difference (p-value: 0.000). This might be because tuberculosis affects the immune system. The A/G ratio, which can show different health conditions, was higher in patients (average: 1.83) compared to controls (average: 1.43), indicating significant changes in protein levels associated with the disease (p-value: 0.000). Additionally, there was a statistically significant difference between the serum calcium levels of the patients (average: 8.95 mg/dl) and the controls (9.49 mg/dl) (p-value: 0.000).

TABLE NO.2: PROTEIN PROFILE COMPARISON PATIENT VS CONTROL

VARIABLES	PATIENTS	CONTROLS	P-VALUE
Total Protein	6.97 ± 0.63	7.26 ± 1.08	0.022*
Albumin	4.91 ± 1.14	4.23 ± 0.52	0.000*
Globulin	2.48 ± 0.63	3.03 ± 0.71	0.000*
A/G Ratio	1.83 ± 0.52	1.43 ± 0.25	0.000*

* Statistically Significant

Discussion

Shingdang J *et al.,* (2016) aims and objectives of the study is to assess the relationship between tuberculosis (TB) and serum proteins, with an emphasis on the albumin/globulin ratio. The results of albumin/globulin ratio was found to be significantly altered between the TB case group and the healthy non T.B control .The albumin levels were raised in cases compared to the control ,while total protein and globulin levels were significantly lower in the study group [14]. This is comparable to the research conducted by **Ramakrishna *et al.*** [20]. which found that TB

patients had lower serum albumin levels than the non-TB control group [21]. According to **Moses *et al.*** (2016), globulin was considerably greater in TB patients while serum total protein and albumin were significantly lower in pulmonary TB patients compared to controls. Hepatotoxicity has been documented in patients taking anti-TB medication, according to earlier research. Serum albumin levels below 3.5 g/L were found to be a risk factor for DIH patients in studies [22] . **Morris *et al.*** reported that hypo albuminemia was present in 72% of the TB patients who were chosen. In comparison to non-TB control groups, our study found that the Test group had lower levels of total proteins, higher levels of serum albumin, lower levels of globulins, and a modest fluctuation in the A/G ratio [23]. **Yakram *et al.*,** (2017) Faisalabad This study compared a number of biochemical and hematological characteristics between patients with tuberculosis (TB) and healthy individuals as well as between TB patients and those who were also co-infected with the hepatitis C virus (HCV). Significantly increased levels of alanine aminotransferase were observed in both the TB control group and the TB + HCV co-infected group. Serum protein concentrations varied significantly amongst the various categories in the population under study. Hemoglobin concentration differed significantly between the co-infected TB + HCV group and the healthy group. [24]. Our study investigated a range of and biochemical parameters in mycobacterium tuberculosis (TB) patients and in those co-infected with hepatitis B and C virus (HCV). These parameters provide valuable insights into the health status of the individuals under study.

Conclusion

Evaluating the findings of current study, a significant decrease in serum Total protein levels in patients compared to controls, Albumin levels were significantly higher in patients than in controls. Patients had lower levels of globulin compared to control, the A/G ratio was higher in patients compared to controls, indicating significant changes in protein levels associated with the disease severity. This study will prove to be restrained with previous works and would acknowledge potential constraints and distinct propositions for advanced investigation in future.

Acknowledgement

None

Authors Contribution

SNM and SMM explored the literature and combined and edited the manuscript and BK,

All authors have contributed substantially and approved the final version of the manuscript.

Funding

Self-expenditure

References

1. Correia-Neves, M., et al., *Biomarkers for tuberculosis: the case for lipoarabinomannan.* ERJ open research, 2019. **5**(1).
2. Koch, A. and V. Mizrahi, *Mycobacterium tuberculosis.* Trends in microbiology, 2018. **26**(6): p. 555-556.
3. Sinha, M., N. Jain, and K. Sudevan, *In silico diagnosis of tuberculosis in multiple species using single nucleotide polymorphism as biological marker.* Int J Adv Tech Eng Sci, 2014. **2**: p. 2348-7550.
4. Mainous III, A.G. and C. Pomeroy, *Management of antimicrobials in infectious diseases*. Vol. 349.S 2001: Springer.
5. Koehler, P.J. and D.J. Lanska, *Neuropathological images in the great pathology atlases.* Journal of the History of the Neurosciences, 2022. **31**(2-3): p. 279-311.
6. Dasaradhan, T., et al., *Tuberculosis-associated anemia: a narrative review.* Cureus, 2022. **14**(8).
7. Abbas, U., et al., *Tuberculosis and diabetes mellitus: Relating immune impact of co-morbidity with challenges in disease management in high burden countries.* Journal of Clinical Tuberculosis and Other Mycobacterial Diseases, 2022. **29**: p. 100343.
8. Rahlwes, K.C., et al., *Pathogenicity and virulence of Mycobacterium tuberculosis.* Virulence, 2023. **14**(1): p. 2150449.
9. Abdelgawad, N., et al., *Linezolid population pharmacokinetic model in plasma and cerebrospinal fluid among patients with tuberculosis meningitis.* The Journal of Infectious Diseases, 2024. **229**(4): p. 1200-1208.
10. Damburam, A., M. Garbati, and H. Yusuph, *Serum proteins in health and in patients with pulmonary tuberculosis in Nigeria.* Journal of Infectious Disease and Immunity, 2012. **4**(2): p. 16-19.
11. Sinha, P., et al., *Serum Protein Electrophoresis Bands As Biomarkers for Drug-Sensitive Pulmonary Tuberculosis.* Cureus, 2023. **15**(8).
12. HKHAN, Z. and S. SWARKE, *Effect of antituberculosis drugs on levels of serum proteins in pulmonary tuberculosis patients.* International Journal of Pharmaceutical Research & Allied Sciences, 2012. **1**: p. 94-100.
13. Shingdang, J., et al., *Serum albumin/globulin ratio in tuberculosis and HIV patients any relationship?* 2016.
14. OKOLIE, C., *Serum Albumin/Globulin ratio in Tuberculosis and HIV Patients any Relationship?* Mycobacterial Diseases, 2016. **6**: p. 199.
15. Powell, S.M., et al., *Longitudinal analysis of host protein serum signatures of treatment and recovery in pulmonary tuberculosis.* Plos one, 2024. **19**(2): p. e0294603.
16. Uchenna, U.C., et al., *Biochemical changes in tuberculosis.* The Pan African Medical Journal, 2021. **38**.

17. Chauhan, A., et al., *Landscaping tuberculosis multimorbidity: findings from a cross-sectional study in India.* BMC Public Health, 2024. **24**(1): p. 453.
18. Fukunaga, R., et al., *Epidemiology of tuberculosis and progress toward meeting global targets—worldwide, 2019.* Morbidity and Mortality Weekly Report, 2021. **70**(12): p. 427.
19. Brockmann, G.A., et al., *Single QTL effects, epistasis, and pleiotropy account for two-thirds of the phenotypic F2 variance of growth and obesity in DU6i x DBA/2 mice.* Genome Research, 2000. **10**(12): p. 1941-1957.
20. Pulimood, A.B., et al., *Differentiation of Crohn's disease from intestinal tuberculosis in India in 2010.* World journal of gastroenterology: WJG, 2011. **17**(4): p. 433.
21. Oveissi, M., A. Nikoonajad, and A. Allami, *Status of Vitamin D, Calcium and Parathyroid Hormone Levels & Hepatorenal Function in Untreated Pulmonary Tuberculosis: A Matched Case-Control Study.* Journal of Advances in Medical and Biomedical Research, 2022. **30**(139): p. 109-115.
22. Moses, A.O., et al., *Assessment of antioxidants and nutritional status of pulmonary tuberculosis patients in Nigeria.* European Journal of General Medicine, 2008. **5**(4).
23. Morris, T.C., et al., *Evaluation of host serum protein biomarkers of tuberculosis in sub-Saharan Africa.* Frontiers in immunology, 2021. **12**: p. 639174.
24. Akram, Y., et al., *Biochemical profiling of tuberculosis patients co-infected with hepatitis C virus.* European Journal of Inflammation, 2017. **15**(1): p. 42-45.