

MONOCYTE LYMPHOCYTE RATIO, PLATELET LYMPHOCYTE RATIO, NEUTROPHIL LYMPHOCYTE RATIO AS PROGNOSTIC MARKERS IN PATIENTS WITH MULTIDRUGS RESISTANT TUBERCULOSIS TREATED AT ULIN GENERAL HOSPITAL BANJARMASIN

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Abstract: *Multi Drugs Resistant Tuberculosis (MDR TB) is tuberculosis with the least resistance to rifampicin and isoniazid. The progresivity and side effects of MDR TB treatment can be monitored through clinical, imaging, and laboratory evaluation. Monocyte Lymphocyte Ratio (MLR), Neutrophil Lymphocyte Ratio (NLR), and Platelet Lymphocyte Ratio (PLR) are the inflammatory markers used as biomarkers of treatment progress in MDR TB. The purpose of this research was to determine the difference of MLR, NLR, and PLR value pre and post MDR TB therapy and to determine the best prognostic biomarker in the evaluation of MDR TB treatment progress at Ulin General Hospital in January-December 2017. This research was an analytic observational study with a cross-sectional design. The sample was 17 patients selected based on the inclusion and exclusion criteria. Results obtained from paired T-tests showed the decreasing of MLR, NLR, PLR value in MDR TB patients receiving TB therapy. PLR had a more role as inflammatory biomarker of MDR TB treatment with $p = 0.000$, compared to MLR ($p = 0.01$), and NLR ($p = 0.006$). The conclusion is PLR was significantly the best prognostic biomarker for MDR TB treatment.*

Keywords: *MDR TB, Monocyte Lymphocyte Ratio, Neutrophil Lymphocyte Ratio, Platelet Lymphocyte Ratio*

INTRODUCTION

Multi Drugs Resistant Tuberculosis (MDR TB) is tuberculosis with the least resistance to rifampicin and isoniazid.¹ Patients with MDR TB are separated from ordinary TB because the prognosis of treatment in MDR TB is worse than the regular TB. In 2014, WHO reported MDR TB incidence worldwide of 480,000 cases with mortality caused by MDR TB reaching 190,000 cases. WHO also said about 300,000 cases of MDR TB found in India, China, and Russia. In Indonesia, WHO estimated MDR TB incidence approximately 9000 new cases by 2014 and Indonesia ranked the 6th place in the world.² There are characteristic of TB and MDR TB patients in Ulin General Hospital Banjarmasin, starting from December 2015 until May 2016.³

The treatment of MDR TB is different from TB treatment without anti-tuberculosis (OAT) drug resistance. Treatment of MDR TB takes a long time (18-24 months) and uses first-line drugs, an injection drug, a fluoroquinolone class drug, and 4th group drug (oral second line), and group 5 drugs as adjuvan. The use of various medications for MDR TB causes some problems regarding tolerance of side effects, as well as monitoring the treatment progress.⁴

The progress and side effects of MDR TB treatment can be observed through clinical examination of patients, imaging study, sputum of Acid Resistant Bacteria (ARB), Xpert Genes, blood test. The blood tests included routine blood (HB, leukocytes, platelets, MCV, MCH, MCHC), diff count (monocytes, lymphocytes, eosinophils, basophils), ESR, SGOT / SGPT, ureum-creatinine, TSH, T3, and fT4.⁴⁻⁷

Changes in monocyte, neutrophil, platelets and lymphocyte values were highly related to inflammatory conditions. Recent studies showed that monocytes lymphocytes ratio (MLR), neutrophils lymphocytes ratio

(NLR), or platelets lymphocyte ratio (PLR) could be used as biomarkers of various diseases other than ESR and CRP.^{7,8} Naess et al. in Norway stated that NLR and MLR value increased in patients with bacterial infections and fever. It also reported that NLR dan MLR were useful predictor value in determining the occurrence of septicemia in patients admitted to hospital due to fever and bacterial infection.⁹ Iqbal *et al.* in Pakistan reported that there was a change in monocyte and lymphocyte values in TB patients before and after receiving TB therapy for 4 months, followed by a progressive decrease in the monocyte-to-lymphocyte ratio (MLR).⁸ Similarly, Qitian et al. in China also reported a significant increase in neutrophils lymphocytes ratio (NLR) and platelet-lymphocyte ratio (PLR) in patients with TB spondylitis which positively correlated with CRP and ESR.⁷

Monitoring the progress of MDR TB treatment through MLR, NLR, and PLR as the prognostic marker of the immunologic response to MDR TB and the alteration of treatment response was essential. In addition, the study of MLR, NLR, and PLR as prognostic markers required more attention. In Ulin General Hospital, there was a research about MDR ATD treatment. It showed that MDR ATD regiment did not cause Anemia and Thrombocytopenia.¹⁰ That research had not showed other Hematology profile, such as MLR, HLR, and PLR which can be used important prognosis marker; thus make this research is also important to be conducted.

RESEARCH METHODS

This was an observational analytic study with a cross-sectional design. This method was intended to assess the influence of the dependent variable on the independent variable at a particular time. Independent variable in this study is MDR TB treatment

with dependent variables were the value of MLR, PLR, NLR before and after MDR TB treatment in Ulin General Hospital Banjarmasin. The population were all TB patients undergoing TB treatment at DOTS clinic at Ulin General Hospital Banjarmasin from January 2017 until December 2017. The sampling used nonprobability method followed by purposive sampling method. The inclusion criteria were MDR TB patients detected by Gene Xpert with positive and negative ARB undergoing MDR TB treatment at Ulin General Hospital for 30 days, aged between 18 years and 76 years, not HIV coinfection, the data and laboratory results are entirely recorded in the medical record. The instrument used in this study was secondary data in the form of medical record and finding of routine blood test and diff count before and after MDR TB treatment in DOTS clinic at Ulin General Hospital Banjarmasin. The procedure in this study was collecting and retrieving data through data register, then traced in the medical record, finally recorded and analysed it. The MLR was derived from the modification of the Iqbal et al. method, by dividing the absolute monocyte value with the absolute lymphocyte value.⁸ The NLR and PLR were obtained in accordance with the definition of PLR and NLR by Abakay et al. NLR was defined as the absolute neutrophil value divided by the absolute lymphocyte value. PLR was defined as the absolute platelets value divided by the absolute lymphocyte value.¹¹

RESULTS AND DISCUSSION

There were 17 samples that participated in this study, consisting of 12 (70.58%) male patients and 5 (29.42%) female patients. The youngest patient was 21 years old, and the oldest was 75 years old with an average age of 47 years old.

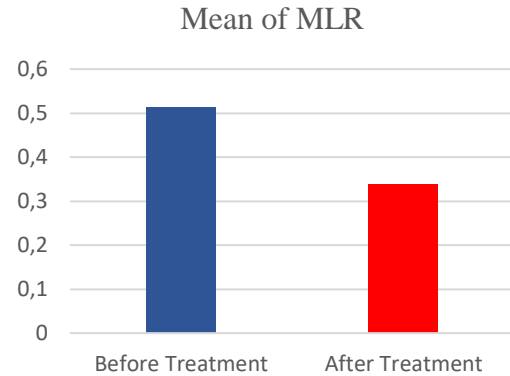


Figure 1. Mean of MLR before and after MDR TB treatment

Pulmonary TB patients had chronic inflammation which altered hematologic their haematology condition. According to Me, 2014, white blood cell changed from leukocytosis to leukopenia, and in some patients were found to be normal.¹² Monocytes and lymphocytes played an essential role in defence against tuberculosis. Monocytes were an essential component of the innate immune response acted as a connector of the adaptive immune system through antigen presentation to lymphocytes. On the other hand, lymphocytes were part of the cellular adaptive immune system which was a significant defence against pulmonary TB. Thus, MLR in peripheral blood circulation represented the capacity of an individual's immune response to pulmonary TB. MLR in some studies was associated with inhibition of *Mycobacterium tuberculosis* (M.tb) growth in vitro, and this MLR was a marker of unknown pathophysiological changes in pulmonary TB.¹³ Wang et al., 2015, reported MLR in TB patients undergoing changes according to anti-TB treatment. Patients with high MLR would decrease, while patients with low MLR would increase. This MLR alteration illustrated the effectiveness of anti-TB therapy.¹³

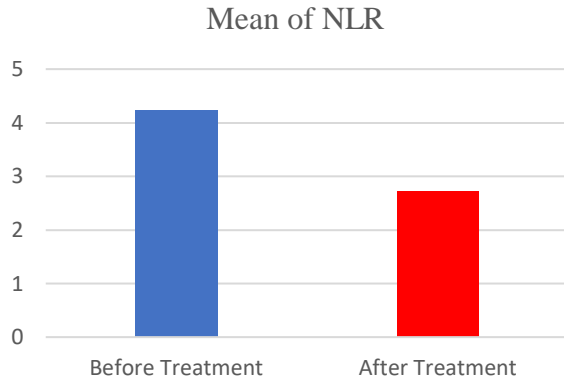


Figure. 2 Mean of NLR before and after MDR TB treatment

Neutrophil altered during pulmonary TB infection. Many studies reported that there was a reverse correlation between pulmonary TB progression and the number of neutrophil in the peripheral blood of active pulmonary TB patients. The less number of neutrophil in vitro caused failure of the immune system to limit the BCG reaction and growth of M.tb.¹⁴ Sutherland *et al.*, 2009 reported that there was an association between the degree severity of pulmonary TB with the number of the neutrophil. In addition, Berry *et al.*, 2010, identified the excessive IFN-inducible gene expression in neutrophil which showed the degree severity of TB, mainly associated with pulmonary tissue destruction.¹⁵ NLR was the ratio of neutrophil to lymphocyte which was used as inflammation marker in some diseases. NLR was a good marker to differentiate pulmonary TB with pulmonary disease caused by other infection. Yoon *et al.*, 2013, stated that NLR was the best marker to diagnose pulmonary TB infection, followed by neutrophil count, diff count, CRP, dan lymphocyte count.¹⁶

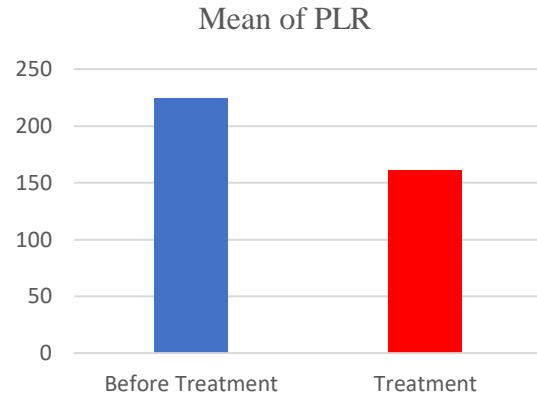


Figure 3. Mean of PLR before and after MDR TB therapy

Figure 3 showed the decreasing of PLR in patients before and after TB MDR therapy. Platelets are known to function as homeostasis in the body. Currently, platelets have been shown to have other functions associated with inflammation and immune responses. As the immune response, platelets produce three granules consisting of dense granules, lysosomes, and alpha granules. The dense granules consist of non-protein small molecules that are important in attracting other platelet responses, e.g. ADP, ATP and serotonin. Serotonin produced from platelets helps neutrophils to stick to the endothelium to get to the site of infection. Platelet lysosomes contain proteases, glycosides, and other bactericidal proteins. Alpha granules produce various cytokines and chemokines that induce natural immunity. In bacterial infections, platelets secrete pro inflammatory mediators and act as guard cells. Reactive thrombocytosis in TB occurred as a part of acute phase reaction and associated with disease severity.¹⁷ PLR is the ratio of platelets to lymphocytes. PLR serves as a prognostic biomarker of various diseases. High PLR indicates an ongoing and unresolved inflammation. Chen *et al.*, 2014, reported that NLR and PLR reflected cell ratio in natural immunity, and both could be used as a marker for differential diagnosis between COPD patients comorbid with TB

infection and COPD patients only. However, Area Under Curve (AUC) PLR was significantly greater than NLR so that PLR might be developed as a biomarker to identify TB infection in COPD patients.¹⁸

Saphiro Wilk test was used to identify the normality of MLR, NLR, and PLR data and got $p > 0.05$. Table 1 showed $n = 0.057$ for MLR, $n = 0.136$ for NLR, and $n = 0.216$ for PLR. It means that all data were normally distributed ($p > 0.05$). After normally distributed data was obtained, the data was tested with the parametric test (paired t-test) to determine the association of MLR, NLR, PLR variable in MDR TB patient before and after therapy.

Table 1. Saphiro Wilk Normality Test

Result		
Saphiro Wilk Test	n	ρ
MLR	17	0,057
NLR	17	0,136
PLR	17	0,216

Figure 4 showed that there was a significant difference of MLR, NLR, and PLR in MDR TB patient before and after therapy (p -value < 0.05). The results of the T test indicated that there was a decrease in the number of MLR, NLR, and PLR in MDR TB patients after receiving MDR TB therapy. It suggested a decrease of inflammation in MDR TB patients who had received treatment. This was in accordance with Iqbal *et al.*, 2014, and Yin *et al.*, 2015, which reported that there was an increase in MLR and NLR in TB patients caused by an inflammatory process that would be impaired after the patient was treated.^{8,20}

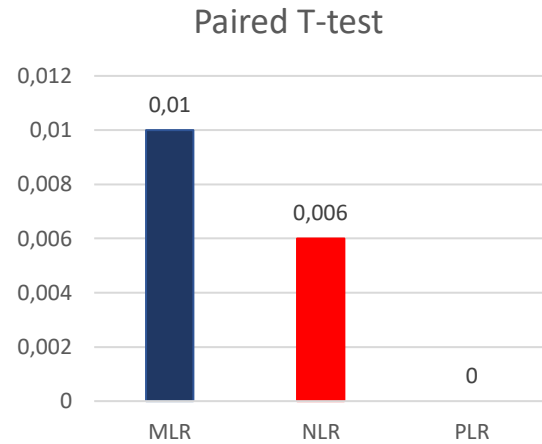


Figure 4. Paired T-test result of MLR, NLR, and PLR in MDR TB

Comparing p value on t test according to table 1, PLR had the smallest p value ($p = 0.000$) compared to MLR ($p = 0.01$) and NLR ($p = 0.006$). This suggested that PLR had a significant change compared to MLR and NLR in MDR TB patients before and after MDR therapy. This was in accordance with Turkmen *et al.*, 2012, which reported that PLR calculations had better inflammatory predictions than NLR in End Stage Renal Disease (ESRD) patients.²¹ Although no similar study has compared the PLR and MLR in various diseases, especially tuberculosis, Naranbhai *et al.*, 2014, stated that MLR could be used as one of the diagnostic markers of TB in TB patients with HIV.²² The report was supported by another study by Naranbhai *et al.*, 2015. It stated that the increasing of monocyte to lymphocytes ratio was a consequence of inflammatory processes mediated by interferon causing an anti-mycobacterial process in infection.²³

There has been no normal standard value of the MLR, NLR, and PLR. Thus, its use limited in determining the marker of inflammation in various diseases. The previous study by Chen *et al.*, 2016, reported that PLR cutoff values > 216.82 and NLR > 2.67 became the gold standard that differentiated between COPD patients only

and COPD patients with pulmonary TB.¹⁹ Other studies by Foget et al., 2017, assigned NLR values of 0.78 to 3.53 in healthy young adults.²⁴ In his study, Ni, 2016, explained that NLR was influenced by age and PLR by age and sex.²⁵ While there has been no cut-off value investigation for MLR. But Stotz, 2014, explained that the MLR value > 2.38 in colon cancer patients was associated with resistance to chemotherapy.²⁶ While Warimwe et al., 2016, reported an association of MLR with the risk of clinical symptoms appearing in falciparum malaria patients.²⁷

CONCLUSION

Our study showed a decrease in MLR, NLR, PLR in MDR TB patients receiving TB therapy. PLR had a better role as a prognostic marker of MDR TB treatment at Ulin General Hospital Banjarmasin with $p = 0.000$, compared with MLR with $p = 0.01$, and NLR with p value = 0.006. Future study will be necessary to validate the function of MLR, NLR, and PLR in MDR TB patients receiving TB therapy.

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