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The Analysis of Predictors of Limb Amputation in Chronic Limb Ischemia (CLI) Patients at Prof. I.G.N.G. Ngoerah Hospital Denpasar

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ABSTRACT

Background: Chronic limb ischemia is a condition where there is an interruption in arterial blood flow that has lasted for 14 days or more. CLI is one of the peripheral arterial diseases with the highest incidence. CLI can progress to a more serious stage and even cause severe morbidity in the form of amputation and death. Although CLI treatment is well developed, the predictors of amputation in CLI patients are not fully understood, especially in Indonesia. Some factors that may be related to the predictors of amputation in patients with CLI are CHF, INR, NLR, ABI, and DVT. ***Objective:*** This study aims to identify predictors of amputation in CLI patients who have received treatment at Prof. I.G.N.G Ngoerah Hospital, Denpasar. ***Method:*** This study used a retrospective cohort research design to determine the predictors of limb amputation in patients with CLI. Data were collected through medical records with a total sample of 46 samples. ***Result:*** The results of bivariate analysis showed that high NLR, ABI, CHF, and DVT had a significant correlation with increased amputation risk in CLI patients. The results of multivariate analysis showed that high CHF and NLR increased the risk of amputation in CLI patients, while type 2 DM, hypertension, and PAD were protective factors, although not statistically significant. ***Conclusion:*** Chronic heart failure and NLR>2 increased amputation risk by 3.6 and 2.8 times, respectively. The use of these predictors may assist clinicians in making treatment decisions to minimize the risk of amputation in CLI patients.

Keywords: Chronic Limb Ischemia (CLI); predictor; amputation; Neutrophil Lymphocyte Ratio (NLR); Ankle Brachial Index (ABI); Deep Vein Thrombosis (DVT).

INTRODUCTION

Peripheral arterial disease is a spectrum of diseases in which blood flow to the limbs is blocked due to the narrowing of the arterial blood vessels. Currently, clinicians and researchers recognize that peripheral arterial disease has similar morbidity and mortality rates as coronary artery disease. (Badimon et al., 2014; Novo et al., 2004). Based on the Global Burden of Disease in 2017, the number of cases of peripheral arterial disease patients in the world in 2017 was 118.1 million cases with an incidence rate of 10.8 million with one-third of cases of symptomatic claudication arising from peripheral arterial disease (Aryaputra et al., 2021).

One of the peripheral arterial diseases with the highest incidence is chronic limb ischemia (CLI). Chronic limb ischemia is a condition where there is an interruption in arterial blood flow that has lasted for 14 days or more.

CLI is characterized by severe pain in the lower limbs, even at rest, and or accompanied by ulcers or gangrene on the skin of the limbs. CLI can progress to a more serious stage, namely critical limb ischemia, which when it occurs can cause severe morbidity in the form of amputation and even death (Bhat et al., 2016; Belaj et al., 2016).

In Indonesia, the incidence of CLI is not known with certainty, but based on the A Global Atherothrombosis Assessment (AGATHA) study by the American Society of Cardiology in 2006, it was found that the incidence of peripheral arterial disease in Indonesia was 9.7%. This number is projected to increase along with the increasing cases of CHF and type 2 diabetes mellitus which are risk factors for peripheral arterial disease, especially CLI (TASC et al., 2015; Nehler et al., 2014; Rollins et al., 2013).

Peripheral arterial disease (PAD) develops faster in patients with diabetes mellitus (DM). The risk of developing CLI is four times higher in patients with DM than in patients without DM. Cardiac comorbidities are common in patients with CLI, and the prevalence of heart failure has been reported to be 10-40% in this population. Platelets and their products are known to play a key role in atherosclerosis. A population-based study in Finland of 888 patients with varicose veins and 2006 control patients found a statistically significant increased likelihood of developing arterial disease (PAD, angina pectoris, Congestive Heart Failure, or cerebrovascular disease) in patients with varicose veins (odds ratio = 2) and suggested that varicose veins and arterial disease may have a common cause. (Chang et al., 2018; Davies, 2012; Khaira et al., 2017; Spreen et al., 2016).

Ankle-brachial index (ABI) <0.9 is sensitive and specific for the diagnosis of PAD and is associated with increased cardiovascular morbidity and mortality, independent of traditional risk factors (Singh et al., 2017). A parameter that is currently being widely investigated for risk stratification and clinical outcomes of CLI patients is the neutrophil-lymphocyte ratio (NLR). The neutrophil-lymphocyte ratio is a marker obtained from a complete blood test. NLR is a simple, quick, and inexpensive test, but a number of studies have shown that NLR values have a strong association with CHF, including peripheral arterial disease. The NLR parameter has also been universally accepted as a predictive index for stent patency, a prognostic factor to determine survival for amputation and mortality in patients with peripheral arterial disease (Luo et al., 2015; Spark et al., 2010).

METHODS

This study used a retrospective cohort research design to determine the predictors of limb amputation in patients with chronic limb ischemia (CLI). Data was taken from the patient's medical record. The study used data on predictors of CHF, ABI, NLR, type 2 DM, and hypertension of CLI patients at the time of diagnosis and the occurrence of amputation within five years after CLI diagnosis. The medical records of CLI patients used were patients diagnosed with CLI within a five-year period from February 2017 to February 2022. Then it was seen within five years after the diagnosis was made whether CLI patients had limb amputation. Samples were taken consecutively using the consecutive sampling method, with a total sample size of 46 samples who met the inclusion criteria.

RESULTS

Characteristics of the study

The basic characteristics of respondents can be seen in Table 1. There were 46 subjects with CLI and from the results of this study, the average age of respondents was 64 ± 11.23 years. The most common gender was female, 57%. The most common comorbidities were type II diabetes mellitus, followed by hypertension, CHF, and renal failure. A total of 25 respondents had a history of smoking and the rest had no history of smoking.

TABLE 1: Basic Characteristics of Research Subjects.

Variable	Not amputated (N=22)	Amputation (N=24)	p-value
Gender			
Female	8 (36%)	18 (75%)	0,008
Male	14 (64%)	6 (25%)	
Mean Age	64 ± 10	64 ± 13	0,909
Type 2 DM			
No	4 (18%)	5 (21%)	0,909
Yes	18 (82%)	19 (79%)	
Hypertension			
No	8 (36%)	9 (38%)	0,090
Yes	14 (64%)	15 (63%)	
CHF			
No	14 (64%)	8 (33%)	0,040
Yes	8 (36%)	16 (67%)	
Renal Failure			
No	12 (55%)	13 (54%)	0,909
Yes	10 (45%)	11 (46%)	
Smoking			
No	13 (59%)	8 (33%)	0,080
Yes	9 (41%)	16 (67%)	
Ulcer			
No	3 (14%)	3 (13%)	0,909
Yes	19 (86%)	21 (88%)	
Fontaine degree			
3	1 (4,5%)	0 (0%)	
4	3 (14%)	1 (4,2%)	0,200
5	18 (82%)	23 (96%)	

Variable	Not amputated (N=22)	Amputation (N=24)	p-value
Rutherford degree			
3	1 (4,5%)	0 (0%)	0,200
4	1 (4,5%)	0 (0%)	
5	3 (14%)	1 (4,2%)	
6	17 (77%)	23 (96%)	
ABI value			
Normal low	2 (9%)	1 (4%)	0,030
Borderline Perfusion	4 (18%)	5 (21%)	
PAD	16 (73%)	18 (75%)	
Mean Leukocyte	14 ± 9	18 ± 7	0,005
Leukocyte categories			
Normal	10 (45%)	3 (13%	0,0013
Leukocytosis	12 (55%)	21 (88%)	
Mean Neutrophil	10,4 ± 9,4	13,4 ± 5,5	0,008
Neutrophils categories			
Normal	11 (50%)	6 (25%)	0,079
High neutrophil (>8)	11 (50%)	18 (75%)	
Mean Lymphocyte	1,74 ± 1,39	1,39 ± 0,64	0,200
Lymphocyte categories			
Normal	15 (68%)	15 (63%)	0,600
Low lymphocyte	6 (27%)	9 (38%)	
High lymphocyte (>4)	1 (4,5%)	0 (0%)	
Mean INR	1,36 ± 1,07	1,25 ± 0,37	0,030
INR categories			
Normal	12 (55%)	14 (58%)	0,400
High INR (>1,1)	10 (45%)	10 (42%)	
Mean NLR	16 ± 17	19 ± 12	0,100
NLR categories			
Normal	6 (27%)	0 (0%)	0,008
High NLR (>2)	16 (73%)	24 (100%)	
DVT			
No	5 (23%)	6 (25%)	0,009
Yes (Score >1,5)	17 (77%)	18 (75%)	

The clinical characteristics of diabetic foot wounds can be seen in Table 1. A total of 40 respondents had ulcers or gangrene. Most of the respondents were already in the final stage based on the Rutherford (87%) and Fontaine (89.1%) classifications. The majority of patients with the PAD category had amputation as many as 14 people. The majority of patients had DVT (76.1%) and as many as 18 or 52% of patients had undergone amputation.

Differences among patients' baseline and clinical characteristics were evaluated by Chi-square test, and data analysis of variables based on the incidence of amputation as shown in Table 5.2 found a significant association between amputation and gender (p=0.008) and CHF factor (p=0.04). Statistical test results with Chi-square also showed a significant relationship between amputation and differences in mean leukocytes (p=0.005), mean neutrophils (p=0.008), and high NLR values (p=0.008).

Factors predictive of amputation in patients with CLI

Differences among baseline and clinical characteristics of patients were evaluated by Chi-square test, data analysis of variables based on the incidence of amputation as shown in Table 2 found a significant association between CHF and amputation in CLI patients (p=0.040). In this study, it was found that ABI had a significant relationship with the incidence of amputation in CLI patients (p=0.030). The results of statistical tests show that high NLR has a significant relationship with amputation in CLI patients (p=0.008). This study found that INR was not significantly associated with amputation in CLI patients (p=0.400). The average INR results in this study were still within normal limits with the administration of anticoagulants such as heparin. The results of this study showed no association between INR and the incidence of amputation. This was attributed to the administration of anticoagulants to patients. The results of this study showed that DVT had a significant association with amputation in CLI patients (p=0.009).

TABLE 2: Bivariate test results of predictors of amputation in CLI patients.

Variable	Not amputated (N=22)	Amputation (N=24)	p-value	OR
CHF				
No	14 (63,6%)	8 (36,4%)		
Yes	8 (33,3%)	16 (66,7%)	0,040	3,500
ABI value				
ABI Borderline Perfusion	2 (66,7%)	1 (33,3%)		
ABI PAD	20 (46,5%)	23 (53,5%)	0,467	2,300
NLR categories				
Normal	12 (66,7%)	6 (33,3%)		
High NLR (>2)	10 (35,7%)	18 (64,3%)	0,040	3,600
Type 2 DM				
No	4 (44,4%)	5 (55,6%)		
Yes	18 (48,6%)	19 (51,4%)	0,559	0,844
Hypertension				
No	8 (47,1%)	9 (52,9%)		
Yes	14 (48,3%)	15 (51,7%)	0,936	0,952

TABLE 3: Multivariate test results CHF, ABI, NLR, Type 2 DM, Hypertension.

Variable	p-value	OR	95%CI	
			Lower bound	Upper bound
CHF	0,088	3,644	0,825	16,098
ABI PAD	0,937	0,896	0,059	13,663
NLR	0,128	2,819	0,742	10,711
Type 2 DM	0,772	0,157	0,157	3,955
Hypertension	0,609	0,154	0,154	2,991

The R2 value generated from this multivariate analysis was 0.203 which means that these variables could only predict the incidence of amputation by 20.3%. The results of this analysis were supported by a P value of 0.179, which means it is not statistically significant. From the multivariate analysis, the variables of CHF and high NLR still had an increased risk of amputation, but other variables including PAD based on ABI, T2DM, and Hypertension failed to show an increased risk of amputation in patients. The presence of CHF increased the risk of amputation up to 3.64 times higher compared to patients without a history of CHF after controlling for the variables of PAD, high NLR, type 2 DM, and hypertension.

However, this OR is not supported by a significant P value of 0.088. This may be due to the relatively small sample size to prove significance. High INR values can also increase the risk of amputation in patients up to 2.82 times higher after controlling for other variables, but it was also not shown to be statistically significant with a P value of 0.128. Other variables such as T2DM, hypertension, and PAD were protective factors in multivariate analysis. However, these protective factors were all not supported by significant P values, so these results cannot be validated and require further research with a larger sample size given that these variables may have a causal effect on the incidence of amputation in patients.

DISCUSSION

Characteristics of Research Subjects

Respondents' Age Characteristics

The results of this study showed that the mean age of respondents was 64 ± 11.73 years. CLI patients are similar to a population that is older on average than non-CLI patients. In one study, more than a quarter were 80 years old or older in this study population. However, almost one-fifth of them were less than 65 years old which is the working-age population. Younger patients had a lower risk of mortality than older patients, but the sex-relative risk ratio at the same age was higher in younger patients. In addition, the incidence of major amputation was higher in the younger population (Takahara M et al., 2020).

Respondents' Gender Characteristics

The most common gender was female at 57%. However, this is different from the study that showed males were more dominant (more than 60%). This may be due to the comorbid influence of the study, which was occupied by type 2 diabetes mellitus.

This study was occupied by type 2 diabetes mellitus. In the case of diabetes mellitus, it is usually dominated by women (Kautzky-Willer et al., 2016).

Based on the results of the analysis, there is a significant difference between the risk of amputation based on gender, with more female patients experiencing amputation than men (p = 0.008). This is different from a study conducted by Mentias et al. in 2020 in America showing a higher incidence of amputation compared to women with a proportion of 2.5% versus 1.9%, and this difference was statistically significant (P = 0.04). In a study conducted by Torbjörnsson et al. in 2019 in Sweden, out of 855 patients with CLI, the risk of amputation in men was slightly higher than in women but statistically, this relationship was not significant (P = 0.028).

Comorbidity Characteristics of Respondents

The most common comorbidities in this study were type 2 DM where as many as 19 people with type 2 DM experienced amputation. Then followed by hypertension where as many as 15 people with hypertension experienced amputation. Patients with comorbid renal failure who experienced amputation were 11 people. Patients with a history of smoking who experienced amputation were 16 people. Research by Rollins et al in 2013 showed that a history of smoking, diabetes mellitus with high HbA1c levels, high non-HDLC levels, and kidney failure, as well as obesity, are known as major accelerators of atherosclerosis, or vascular aging (Rollins et al., 2013).

The results of several studies show that patients with DM are at four times greater risk of developing CLI than patients without DM. This is because patients with DM often have peripheral neuropathy with sensory dysfunction which is thought to contribute to lower limb ulcers and progressive tissue loss in patients with CLI. The Rutherford and Fontaine classification were originally created to help clinicians make amputation decisions and predict patient outcomes. Amputation in the PAD population is usually performed in the case of CLI. A study showed data that there has been a decrease in the rate of major amputations in the CLI population over the last 2 decades since the use of this classification (Levin *et al.*, 2019).

Predictors of CHF with amputation in patients with CLI

This study found that CHF had a significant association with the incidence of amputation in CLI patients (p=0.040). Patients with patients with comorbid CHF 3.5 times increased the incidence of amputation in CLI patients (OR: 3,500; 95% CI 1,039-11,789). The results of another study of heart failure in post-randomization patients with major amputation showed an association with an incidence rate of 34 patients (15.7%) of the total sample (Long et al., 2020). The decreased or low level of tissue perfusion in CHF has an impact on the decrease in oxygen present in the tissues, especially the extremities.

This study also showed a significant difference between CLI patients with CHF and those without CHF, patients with CHF risk experienced more amputations. The results of this study are similar to research conducted by Nishijima et al. 2017 in Japan which showed from 129 patients with CLI, there were 83 patients (69%) patients with CHF disorder, and 82% of this group underwent major amputation, and 63% of this group underwent minor amputation. The incidence of CHF was higher in patients with major amputation (P = 0.042).

Another study by Constantinescu et al. (2013) also showed an association between CHF and amputation in CLI showing decompensated congestive heart failure seems to be more prone to unfavorable progression of minor or major amputations performed below the knee. This may be due to more severe atherosclerotic involvement, making minor amputations insufficient. Severe cardiac involvement may also lead to low cardiac output, with insufficient blood supply to the lower extremities, or may lead to acute ischemia of the atherosclerotic field through emboligenic mechanisms.

Predictors of ABI with Amputation in Patients with CLI

In this study, it was found that ABI had a significant association with the incidence of amputation in CLI patients (p=0.030). Patients with the "Borderline Perfusion" category 1.34 times increased the incidence of amputation in CLI patients (OR: 1,340; 95% CI 1,161-18,624), and the "PAD" category 1.5 times increased the incidence of amputation in patients (OR: 1,500; 95% CI 1,271-31,575). Research by Paskiewicz et al in 2021 showed the relationship between ABI and amputation in CLI, namely individuals with low ABI (≤0.90), low ABI at the threshold (0.91-1.00), and ABI 1.01 to 1.10 times increased the occurrence of severe leg CLI and ischemic leg amputation compared to people with normal ABI (1.11-1.20) (Paskiewicz et al., 2021).

An elevated or unmeasured ABI was shown to be an independent predictor of major amputation. In these cases, the ABI is artificially raised as higher cuff pressure is required to compress the stiff artery. The artery may not even be compressible at all, resulting in an unmeasurable ABI. Previous studies looked at the prognostic value of ABI for heart disease and mortality in high-risk populations. Its predictive value for established CLI has been less studied. Three externally validated prediction models focusing on CLI patients did not include ABI. Only the Project Engineering Ex-Vivo Venous Grafts by Transfection III (PREVENT III) model reported amputation-free survival (AFS), the most important clinical outcome parameter for CLI. Ninety-five percent of patients with high/unmeasured ABI underwent major amputation or died within 5 years. A high/unmeasurable ABI indicates a poorly compressible or incompressible artery in the lower extremities (Spreen et al., 2018).

ABI values (and corresponding ankle pressures) are useful for predicting the limb output of CLI patients. Ankle pressures <50 mm Hg are associated with a higher risk of amputation.

An increased risk of amputation has been reported in ABI values <0.50 in non-revascularized patients with foot ulcers. An ABI of 0.90 is strongly associated (odds ratio: 8.2) with a 7-year amputation risk in people with diabetes mellitus. Some studies report greater accuracy of ankle pressure, rather than ABI, for predicting extremity clinical prognosis (Marston et al., 2006).

Predictors of NLR with Amputation in Patients with CLI

Statistical test results showed that high NLR had a significant association with amputation in CLI patients (p=0.008). Patients with high NLR 2.5 times increase the incidence of amputation in CLI patients (OR: 2.500; 95% CI 1.710-3.654). Another study regarding the relationship between NLR and amputation was proposed by (Ye et al., 2020) which showed the results of NLR having a positive relationship with the severity of lower extremity peripheral arterial disease and having a higher ratio related to poor prognosis, especially the risk of re-hospitalization for one year. Higher NLR also correlated with mortality within one year.

Another study by (Luo et al., 2015) showed NLR is an independent predictive factor for amputation in critical limb ischemia patients. Patients with a post-treatment neutrophil-lymphocyte ratio ≥3.8 were most likely to undergo amputation; amputation-free survival usually occurred in patients with a post-treatment neutrophil-lymphocyte ratio <3.8. CLI is one type of long-term atherosclerosis, and the development of fibrosis is often associated with an immune inflammatory response. Fibrosis, which aggravates ischemia, is caused by recurrent local inflammatory responses involving neutrophils and lymphocytes. NLR is a good systematic inflammatory marker that indicates inflammatory disease. NLR predicts late events in other medical fields, such as myocardial ischemia and severe sepsis. When severe ischemia occurs and infection and immune disease are not ruled out simultaneously, a systematic increase in NLR post-NLR may indicate an irreversible ischemic condition that can lead to late events, such as amputation. Another study by (Mughal et al., 2019) also showed high NLR values were associated with a lower one-year survival rate (53%) than low NLR values (78%).

Another study showed preoperative NLR values > 3.95 were associated with primary patency failure within 12 months (16.47% vs. 89.21%; p < 0.0001), with higher amputation rates (42.35% vs. 2.88%; p < 0.0001), and with higher mortality rates (27.06% vs. 2.88%; p < 0.0001). Moreover, according to ROC analysis, depending on the Rutherford classification, NLR values > 3.95 and PLR values > 142.13 were associated with all significant adverse events, except death in RC 2, and amputation and mortality in RC 5. NLR may increase clinical susceptibility to poor outcomes after revascularization, making it the first line in a series of predictable biochemical surveillance (Russu et al., 2022).

Predictors of Type 2 Diabetes Mellitus with amputation in patients with CLI

In this study, diabetes mellitus was not associated with amputation in patients with CLI with an odds ratio of 0.788 and a confidence interval of 1.577 - 0.3955. Although in theory diabetes mellitus is associated with amputation in PAD patients. This may be because the patient's blood sugar is already controlled. Costa, et al. showed that the highest prevalence of amputation in patients with diabetes mellitus was found in the elderly, especially in the group aged more than 60 years, while in this study the mean age was 64 + 10 years and 64 + 13 years in the non-amputated and amputated groups, but the study showed that the increase in amputation rates in diabetics was seen in patients with other comorbidities such as physical inactivity, smoking, heart disease, stroke, chronic renal failure, and diabetic foot, while in this study other comorbidities were not assessed (Aryaputra et al., 2023; Costa et al., 2020).

A study showed that T2DM duration > 20 years, hypertension, and diabetic ulcer with Wagener classification > 4 were predictors of amputation in T2DM patients. Diabetic ulcer is the most severe and common complication and the main cause of hospitalization. Some studies have shown that T2DM can increase the risk of lower limb amputation up to 20-fold. However, a study showed that there are other factors in patients with diabetes mellitus that are associated with amputation incidence such as duration of T2DM. In addition, the presence of nephropathy or neuropathy. T2DM patients with hypertension are also associated with faster microvascular and macrovascular complications. Treatment of T2DM is also related to the amputation risk of patients with T2DM (Nouira et al., 2023; Zerovsnik et al., 2022).

Predictors of Hypertension with Amputation in Patients with CLI

Hypertension can cause arteries to stiffen as it can affect the release of vasoactive factors from the endothelium and adversely affect systemic arterial vessels resulting in lower extremity arterial disease. Peripheral arterial disease is a form of atherosclerosis that mostly occurs in the lower extremities. Hypertension can cause blood vessels to stiffen due to integrin activation and talin recruitment resulting in the formation of focal adhesion complexes and subsequent signaling pathways. The findings of this study showed that hypertension had no impact on the incidence of amputation of CLI patients with an odds ratio of 0.679 and a confidence interval of 0.1544 - 2.991. This finding is in contrast to the study by Pinto et al which showed that PAD patients who underwent amputation tended to have high levels of arterial stiffness compared to those who did not undergo amputation. This difference may be due to the administration of hypertension drugs such as calcium channel blockers (CCBs) that can significantly improve peripheral arterial disease (Mendes-Pinto et al., 2021; Xu et al., 2022).

CONCLUSIONS

Based on the analysis of demographic characteristics, most of the respondents were female (57%). The mean age of respondents was 64 ± 11.23 years with the most comorbid diseases. Congestive Heart Failure (CHF) and NLR > 2 increased amputation risk 3.6 and 2.8 times higher respectively but ABI, type 2 DM, and hypertension did not increase amputation risk in CLI patients.

REFERENCES

[1] Abraham, A.T., Mojaddedi, S., Loseke, I.H., Bray, C., 2024. Hypertension in Patients with Peripheral Artery Disease: An Updated Literature Review. Cureus 16, e62246. https://doi.org/10.7759/cureus.62246

[2] American Diabetes Association Professional Practice Committee, 2022. 6. Glycemic Targets: *Standards of Medical Care in Diabetes—2022*. Diabetes Care 45, S83–S96. https://doi.org/10.2337/dc22-S006

[3] Arbănași, E. M., Mureșan, A. V., Coșarcă, C. M., Kaller, R., Bud, T. I., Hosu, I., Voidăzan, S. T., Arbănași, E. M., & Russu, E. (2022). Neutrophil-to- Lymphocyte Ratio and Platelet-to-Lymphocyte Ratio Impact on Predicting Outcomes in Patients with Acute Limb Ischemia. *Life (Basel, Switzerland)*, *12*(6), 822. https://doi.org/10.3390/life12060822

[4] Aryaputra, A.B., Bagaswoto, H.P., Nugroho, T., Fauziyati, A., 2023. Acute Limb Ischemia on Diabetic Patient: A Case Report and Literature Review. Proceedings of the 3rd International Conference on Cardiovascular Diseases (ICCvD 2021) 161–172. https://doi.org/10.2991/978-94-6463-048-0_19

[5] Badimon, Lina, dan Gemma Vilahur. 2014. Thrombosis formation on atherosclerotic lesions and plaque rupture. *Journal of internal medicine* 276.6: 618-632.

[6] Belaj, Klara, *et al.* 2016. Association of the derived neutrophil–lymphocyte ratio with critical limb ischemia. *Angiology*. 67.4: 350-354.

[7] Bhat, Tariq M., Maxwell E. Afari, dan Lawrence A. Garcia. 2016. Neutrophil lymphocyte ratio in peripheral vascular disease: a review. *Expert review of cardiovascular therapy*.14.7: 871-875.

[8] Buonacera, A., Stancanelli, B., Colaci, M., & Malatino, L. 2022. Neutrophil to Lymphocyte Ratio: An Emerging Marker of the Relationships between the Immune System and Diseases. In *International Journal of Molecular Sciences* (Vol. 23, Issue 7). MDPI. https://doi.org/10.3390/ijms23073636

[9] Chang, Leun Shyue, *et al.* 2018. Association of Varicose Veins with Incident Venous Thromboembolism and Peripheral Artery Disease. *JAMA*. 2018;319(8):807-817. doi:10.1001/jama.2018.0246

[10] Costa, W.J.T., Penha-Silva, N., Bezerra, I.M.P., Paulo Dos Santos, I., Ramos, J.L.S., Castro, J.M.D., Pereira, J.E.G., Silva, A.P.D., Monteiro, A., De Abreu, L.C., 2020. Analysis of Diabetes Mellitus-Related Amputations in the State of Espírito Santo, Brazil. Medicina 56, 287. https://doi.org/10.3390/medicina56060287

[11] Cronenwett, Jack L., dan K. Wayne Johnston. 2014. Rutherford's vascular surgery e-book. *Elsevier Health Sciences*.

[12] Constantinescu, I., Petru Constantinescu, D., Chiș, B., Andercou, A., & Mironiuc,I. A. 2013. Influence of rIsk factors and comorbIdItIes on the successful therapy and survIval of patients wIth critical limb Ischemia. *Original Research Clujul Medical*, *86*(1).

[13] Davis, M.G. 2012. Critical limb ischemia: epidemiology. *Methodist DeBakey Heart & Vascular Center*. VIII (4) 2012.

[14] Davies, M. G. 2012. *CritiCial liMB isCheMia: ePideMioloGy*. *8*(4).

[15] Earnshaw, Jonothan J. 2010. Acute ischaemia: Evaluation and decision making. Rutherfords Vascular Surgery. 7th ed. Philadelphia, *PA: Saunders Elsevier*: 2389-2398.

[16] Farber, A., 2018. Chronic Limb-Threatening Ischemia. N Engl J Med 379, 171–180. https://doi.org/10.1056/NEJMcp1709326

[17] Fei, Y. F., *et al.* 2012. Incidence and risk factors of amputation among inpatients with diabetic foot. *Zhonghua Yi Xue Za Zhi*. 92.24: 1686-1689.

[18] Gary, Thomas, *et al.* 2013. Neutrophil-to-lymphocyte ratio and its association with critical limb ischemia in PAOD patients. *PLoS One* 8.2: e56745.

[19] Girolami, Bruno, *et al.* 1999. Treatment of intermittent claudication with physical training, smoking cessation, pentoxifylline, or nafronyl: a meta- analysis. *Archives of Internal Medicine*. 159.4: 337-345.

[20] Gonzalez-Fajardo, Jose A., *et al.* 2014. Prognostic significance of an elevated neutrophil–lymphocyte ratio in the amputation-free survival of patients with chronic critical limb ischemia. *Annals of vascular surgery*. 28.4: 999-1004.

[21] Gottsäter, Anders. 2006. Managing risk factors for atherosclerosis in critical limb ischaemia. *European journal of vascular and endovascular surgery*. 32.5: 478-483.

[22] Harrison, D.G., Coffman, T.M., Wilcox, C.S., 2021. Pathophysiology of Hypertension: The Mosaic Theory and Beyond. Circulation Research 128, 847–863. https://doi.org/10.1161/CIRCRESAHA.121.318082

[23] Henry, Antonia J., *et al.* 2011. Socioeconomic and hospital-related predictors of amputation for critical limb ischemia. *Journal of vascular surgery* 53.2: 330-339.

[24] Herlihy, D. R., Thomas, M., Tran, Q. H., & Puttaswamy, V. (2020). Primary prophylaxis for venous thromboembolism in people undergoing major amputation of the lower extremity. *The Cochrane database of systematic reviews*, *7*(7),CD010525. https://doi.org/10.1002/14651858.CD010525.pub3

[25] Hinchliffe, Robert J., *et al.* 2016. Effectiveness of revascularization of the ulcerated foot in patients with diabetes and peripheral artery disease: a systematic review. *Diabetes/metabolism research and reviews*. 32: 136-144. Hopper, I., & Easton, K. 2017. Chronic heart failure. *Australian Prescriber*, *40*(4), 128–136. https://doi.org/10.18773/austprescr.2017.044

[26] Jakubiak, G.K., Pawlas, N., Cieślar, G., Stanek, A., 2020. Chronic Lower Extremity Ischemia and Its Association with the Frailty Syndrome in Patients with Diabetes. IJERPH17, 9339. https://doi.org/10.3390/ijerph17249339

[27] Huang, Li Hsuang, *et al.* 2019. Risk stratification for low extremity amputation in critical limb ischemia patients who have undergone endovascular revascularization: A survival tree analysis. *Medicine* (2019) 98:33(e16809).

[28] Kautzky-Willer, A., Harreiter, J., & Pacini, G. (2016). Sex and Gender Differences in Risk, Pathophysiology and Complications of Type 2 Diabetes

[29] Mellitus. *Endocrine reviews*, *37*(3),278–316. https://doi.org/10.1210/er.2015-1137

[30] Khaira, *et al.* 2017. Long-term outcomes in patients with critical limb ischemia and heart failure with preserved or reduced ejection fraction. *Vascular Medicine* 2017, Vol. 22(4) 307–315

[31] Kullo, I. J., & Rooke, T. W. (2016). CLINICAL PRACTICE. Peripheral Artery Disease. *The New England journal of medicine*, *374*(9), 861–871. https://doi.org/10.1056/NEJMcp1507631

[32] Lin, Jonathan H., *et al.* 2019. Endovascular-first treatment is associated with improved amputation-free survival in patients with critical limb ischemia. Circulation: *Cardiovascular Quality and Outcomes* 12.8: e005273.

[33] Levin, Scott R.; Arinze, Nkiruka; Siracuse, Jeffrey J. (2019). *Lower Extremity Critical Limb Ischemia: A Review of Clinical Features and Management. Trends in Cardiovascular Medicine,(), S1050173819300477–.* doi:10.1016/j.tcm.2019.04.002

[34] Long, C. A., Mulder, H., Fowkes, F. G. R., Baumgartner, I., Berger, J. S., Katona, B. G., Mahaffey, K. W., Norgren, L., Blomster, J. I., Rockhold, F. W., Hiatt, W. R., Patel, M. R., Jones, W. S., & Nehler, M. R. 2020. Incidence and Factors Associated with Major Amputation in Patients with Peripheral Artery Disease: Insights from the EUCLID Trial. *Circulation: Cardiovascular Quality and Outcomes*, *13*(7), 395–403. https://doi.org/10.1161/CIRCOUTCOMES.119.006399

[35] Luo, Han, *et al.* 2015. Post-treatment neutrophil-lymphocyte ratio independently predicts amputation in critical limb ischemia without operation. *Clinics* 70: 273-277.

[36] Made, Oka Sastrawan, Putu Yasa Ketut, dan Raka Widiana I. Gede. 2019. Faktor- faktor risiko yang berhubungan dengan amputasi paska intervensi endovaskular pada penderita peripheral arterial disease rutherford 2–4 di RSUP Prof. I.G.N.G. Ngoerah Denpasar. *Medicina* 50.1: 214-220.

[37] Mendes-Pinto, D., Rodrigues-Machado, M.D.G., Avelar, G.L., Navarro, T.P., Dardik, A., 2021. Arterial stiffness predicts amputation and death in patients with chronic limb-threatening ischemia. Journal of Vascular Surgery 74, 2014-2022.e4. https://doi.org/10.1016/j.jvs.2021.05.052

[38] Mentias, A., Vaughan-Sarrazin, M., Saad, M. and Girotra, S., 2020. Sex differences in management and outcomes of critical limb ischemia in the Medicare population. Circulation: *Cardiovascular Interventions,* 13(10), p.e009459.

[39] Modi, S., Deisler, R., Gozel, K., Reicks, P., Irwin, E., Brunsvold, M., Banton, K., & Beilman, G. J. (2016). Wells criteria for DVT is a reliable clinical tool to assess the risk of deep venous thrombosis in trauma patients. *World journal of emergency surgery : WJES*, *11*, 24. https://doi.org/10.1186/s13017-016- 0078-1

[40] Mughal, N. A., Fagan, A., Atkins, E. R., Awopetu, A., & Coughlin, P. 2019. The Neutrophil–Lymphocyte Ratio is a Predictor of 1-Year Survival Following Major Limb Amputation. *European Journal of Vascular and Endovascular Surgery*, *58*(6).

[41] Nehler, Mark R., *et al.* 2014. Epidemiology of peripheral arterial disease and critical limb ischemia in an insured national population. *Journal of vascular surgery* 60.3: 686-695.

[42] Nishijima, A., Yamamoto, N., Yoshida, R., Hozawa, K., Yanagibayashi, S., Takikawa, M., et al. 2017. Coronary artery disease in patients with critical limb ischemia undergoing major amputation or not. Plastic and Reconstructive Surgery Global Open, 5(6).

[43] Nouira, S., Ach, T., Bellazreg, F., Ben Abdelkrim, A., 2023. Predictive Factors for Lower Limb Amputation in Type 2 Diabetics. Cureus 15, e39887. https://doi.org/10.7759/cureus.39987

[44] Novo, S., G. Coppola, dan G. Milio. 2004. Critical limb ischemia: definition and natural history. *Current Drug Targets-Cardiovascular & Hematological Disorders* 4.3: 219-225.

[45] Paskiewicz, A., Wang, F. M., Yang, C., Ballew, S. H., Kalbaugh, C. A., Selvin, E., Salameh, M., Heiss, G., Coresh, J., & Matsushita, K. 2021. Ankle-brachial index and subsequent risk of severe ischemic leg outcomes: The aric study. *Journal of the American Heart Association*, *10*(22). https://doi.org/10.1161/JAHA.121.021801

[46] Pasqui, Edoardo, *et al.* 2021. The relation between neutrophil/lymphocyte and platelet/lymphocyte ratios with mortality and limb amputation after acute limb ischaemia. *Vascular*: 17085381211010012.

[47] Paquissi, Feliciano Chanana. 2016. The role of inflammation in cardiovascular diseases: the predictive value of neutrophil–lymphocyte ratio as a marker in peripheral arterial disease. *Therapeutics and clinical risk management.* 12: 851.

[48] Rollins, K. E., D. Jackson, dan P. A. Coughlin. 2013. Meta-analysis of contemporary short-and long-term mortality rates in patients diagnosed with critical leg ischaemia. *Journal of British Surgery* 100.8: 1002-1008.

[49] Russu, E., Mureşan, A. V., Arbănaşi, E. M., Kaller, R., Hosu, I., Voidăzan, S., Arbănaşi, E. M., & Coşarcă, C. M. 2022. The Predictive Role of NLR and PLR in Outcome and Patency of Lower Limb Revascularization in Patients with Femoropopliteal Disease. *Journal of Clinical Medicine*, *11*(9). https://doi.org/10.3390/jcm11092620

[50] Santilli, Jamie D., dan Steven M. Santilli. 1999. Chronic critical limb ischemia: diagnosis, treatment and prognosis. American family physician 59.7:1899.

[51] Sastroasmoro, Sudigdo, dan Sofyan Ismael. 2011. Dasar-dasar metodologi penelitian klinis Edisi ke-4. Jakarta: *Sagung Seto* 376.

[52] Schanzer, Andres, *et al.* 2008. Risk stratification in critical limb ischemia: derivation and validation of a model to predict amputation-free survival using multicenter surgical outcomes data. *Journal of vascular surgery*. 48.6: 1464-1471.

[53] Simon, F., *et al.* 2018. Pathophysiology of chronic limb ischemia. *Gefässchirurgie.* 23.1: 13-18.

[54] Singh, GD., *et al.* Non-compressible ABIs are associated with an increased risk of major amputation and major adverse cardiovascular events in patients with critical limb ischemia. *Vascular Medicine* 1–8. https://doi.org/10.1177/1358863X16689831

[55] Spark, James Ian, *et al.* 2010. An elevated neutrophil-lymphocyte ratio independently predicts mortality in chronic critical limb ischemia. *Journal of vascular surgery*. 52.3: 632-636.

[56] Spreen, M.I, *et al.* 2016. Diabetes Mellitus Is Associated with Decreased Limb Survival in Patients with Critical Limb Ischemia: Pooled Data from Two Randomized Controlled Trials. *Diabetes Care*. DOI: 10.2337/dc16-0850

[57] Spreen, M. I., Gremmels, H., Teraa, M., Sprengers, R. W., Martens, J. M., Verhaar, M. C., Wever, J. J., de Borst, G. J., Vos, J. A., Mali, W. P. T. M., & van Overhagen, H. 2018. High and immeasurable ankle-brachial index as predictor of poor amputation-free survival in critical limb ischemia. *Journal of Vascular Surgery*, *67*(6), 1864-1871.e3. https://doi.org/10.1016/j.jvs.2017.10.061

[58] Takahara M, Iida O, Soga Y, Kodama A, Terashi H, Suzuki K, Sugimoto I, Azuma N. Heterogeneity of Age and Its Associated Features in Patients with Critical Limb Ischemia. Ann Vasc Dis. 2020 Sep 25;13(3):300-307. doi: 10.3400/avd.oa.20-00067. PMID: 33384734; PMCID: PMC7751078.

[59] Taşoğlu, Irfan, *et al.* 2014. Usefulness of neutrophil/lymphocyte ratio as a predictor of amputation after embolectomy for acute limb ischemia. *Annals of vascular surgery*. 28.3: 606-613.

[60] Taurino, M., Aloisi, F., Del Porto, F., Nespola, M., Dezi, T., Pranteda, C., Rizzo, L., & Sirignano, P. (2021). Neutrophil-to-Lymphocyte Ratio Could Predict Outcome in Patients Presenting with Acute Limb Ischemia. *Journal of clinical medicine*, *10*(19), 4343. https://doi.org/10.3390/jcm10194343

[61] Teperman, Jacob, *et al.* 2017. Relationship between neutrophil-lymphocyte ratio and severity of lower extremity peripheral artery disease. *International journal of cardiology*. 228: 201-204.

[62] Teraa, M., Conte, M. S., Moll, F. L., & Verhaar, M. C. (2016). Critical Limb Ischemia: Current Trends and Future Directions. *Journal of the American HeartAssociation*, *5*(2),e002938. https://doi.org/10.1161/JAHA.115.002938

[63] TASC Steering Committee*, *et al.* 2015. An update on methods for revascularization and expansion of the TASC lesion classification to include below-the-knee arteries: a supplement to the Inter-Society Consensus for the Management of Peripheral Arterial Disease (TASC II). *Vascular Medicine* 20.5: 465-478.

[64] Tendera M. 2011. ESC Guidelines on The Diagnosis and Treatment of Peripheral Artery Disease. Poland: *The European Society of Cardiology*.

[65] Torbjörnsson, E., Blomgren, L., Fagerdahl, A.M., Boström, L., Ottosson, C. and Malmstedt, J., 2020. Risk factors for amputation are influenced by competing risk of death in patients with critical limb ischemia. *Journal of Vascular Surgery*, *71*(4), pp.1305-1314.

[66] Unger, T., Borghi, C., Charchar, F., Khan, N.A., Poulter, N.R., Prabhakaran, D., Ramirez, A., Schlaich, M., Stergiou, G.S., Tomaszewski, M., Wainford, R.D., Williams, B., Schutte, A.E., 2020. 2020 International Society of Hypertension Global Hypertension Practice Guidelines. Hypertension 75, 1334–1357.

[67] Silvestre J-S and Lévy BI. (2006). Molecular Basis of Angiopathy in Diabetes Mellitus. Circulation Research 98(1). American Heart Association: 4–6. DOI: 10.1161/01.RES.0000200396.90220.41.

[68] Virani SS, Alonso A, Benjamin EJ, Bittencourt MS, Callaway CW, Carson AP, et al. Heart disease and stroke statistics—2020 update: a report from the American Heart Association.

[69] Wang, Qi, *et al.* 2017. Neutrophil-to-lymphocyte ratio is effective prognostic indicator for post-amputation patients with critical limb ischemia. *Saudi medical journal.* 38.1: 24.

[70] Williams, D.M., Jones, H., Stephens, J.W., 2022. Personalized Type 2 Diabetes Management: An Update on Recent Advances and Recommendations. DMSO Volume 15, 281–295. https://doi.org/10.2147/DMSO.S331654

[71] Xu, D., Zhu, X., Huo, J., Xie, X., Huang, C., Fang, X., Yin, T., 2022. A

[72] Nomogram for Predicting the Risk of Critical Limb Ischemia in Adults with Hypertension: A Retrospective Study. IJGM Volume 15, 8205–8216. https://doi.org/10.2147/IJGM.S342448

[73] Xu, Y., Zhang, W., He, J., Wang, Y., Chen, R., Shi, W., 2022. Nomogram for predicting overall survival in patients with triple-negative apocrine breast cancer: Surveillance, epidemiology, and end results-based analysis. The Breast 66.

[74] Ye, M., Qian, X., Guo, X., Wang, H., Ni, Q., Zhao, Y., Xue, G., Deng, H., & Zhang, L. 2020. Neutrophil-Lymphocyte Ratio and Platelet-Lymphocyte Ratio Predict Severity and Prognosis of Lower Limb Arteriosclerosis Obliterans. *Annals of Vascular Surgery*, *64*, 221–227. https://doi.org/10.1016/j.avsg.2019.09.005

[75] Ying, A.F., Tang, T.Y., Jin, A., Chong, T.T., Hausenloy, D.J., Koh, W.-P., 2022. Diabetes and other vascular risk factors in association with the risk of lower extremity amputation in chronic limb-threatening ischemia: a prospective cohort study. Cardiovasc Diabetol 21, 7. https://doi.org/10.1186/s12933- 021-01441-0

[76] Zerovnik, S., Kos, M., Locatelli, I., 2022. Risk of lower extremity amputations in patients with type 2 diabetes using sodium-glucose co-transporter 2 inhibitors. Acta Diabetol 59, 233–241. https://doi.org/10.1007/s00592-021-01805-8

[77] Zwicky, S., F. Mahler, dan I. Baumgartner. 2002. Evaluation of clinical tests to assess perfusion in chronic critical limb ischemia. VASA. *Zeitschrift fur Gefasskrankheiten* 31.3: 173-178.