ONKO RESEARCH
ISSN Electronic version

ORIGINAL ARTICLE

Neutrophil-to-lymphocyte Ratio as a Prognostic Biomarker in Peruvian Patients with Acral Melanoma

Indice Neutrófilo-Linfocito como Biomarcador Pronóstico en Pacientes Peruanos con Melanoma Acral

Julio Maraví[1,a], Anais Cámara[2,a], Sally Paredes[3,4,a], Brady Beltrán[3,4,a,b], Denisse Castro-Uriol[3,4,a]

[1] Unidad Funcional de Oncología Médica, Hospital Nacional Hipólito Unanue, Lima, Perú.
[2] Centro Oncológico ALIADA, Lima, Perú.
[3] Departamento de Oncología y Radioterapia, Hospital Nacional Edgardo Rebagliati Martins, Lima, Perú.
[4] Centro de Investigación de Medicina de Precisión, Universidad de San Martin de Porres, Lima, Perú.
[a] MD, [b] MSc

OPEN ACCES

Cite as:
Maraví J, Cámara A, Paredes S, Beltrán B, Castro-Uriol D. Neutrophil-to-lymphocyte ratio as a prognostic biomarker in acral melanoma: a retrospective single-cohort study in Peru. Onkoresearch. 2024;2(1).

Received:
20-03-2024

Approved:
03-05-2024

Author's contributions:
MJ, CA, PS, BB, and CD participated in the conception of the article, discussion of evidence, writing and approval of the final version.

Financing:
None.

Conflicts of interest:
The authors declare that they have no conflict of interest.

Correspondence:
Julio Maraví
E-mail: Julioeduardomr@outlook.es

ABSTRACT

Objective. Our aim was to evaluate whether NLR, LMR, and PLR serve as prognostic biomarkers in AM, the most frequent subtype of cutaneous melanoma (CM) in Peru. **Materials and methods.** A retrospective study was conducted, including patients diagnosed with AM between 2010 and 2015. Survival analysis was performed using Kaplan-Meier curves and comparisons were made using the log-rank test. Univariate and multivariate survival models were constructed using Cox regression analysis. A p-value less than 0.05 was considered significant. **Results.** Among our cohort of 135 patients with CM, 51.1% (69 cases) had AM. The median age was 68 years, with a predominance of females (55%), and 88.4% had the plantar site as the primary site. The 5-year overall survival (OS) rate was 54.3%. In the univariate analysis, Clark level III/IV, anaplasia, lymphocytic invasion, stage III-IV, and NLR were associated with poor prognosis. In multivariate analysis, NLR >3.5 (HR 3.9, 95% CI 1.5-10.3, p=0.005) and Clark level III-IV (HR 3.5, 95% CI 1.6-7.8, p=0.002) were associated with poor OS. **Conclusion.** NLR emerges as an independent prognostic factor for OS among Peruvian patients with AM in a single cancer center institution.

Keywords
Melanoma, cutaneous malignant; prognostic factor; survival (source: MeSH-NLM).

RESUMEN

Objetivo. Nuestro objetivo fue evaluar si el NLR, LMR y PLR sirven como biomarcadores pronósticos en el melanoma acral (MA), el subtipo más frecuente de melanoma cutáneo (CM) en Perú. **Materiales y métodos.** Se realizó un estudio retrospectivo que incluyó pacientes diagnosticados con MA entre 2010 y 2015. Se realizó un análisis de supervivencia utilizando curvas de Kaplan-Meier y se realizaron comparaciones utilizando la prueba de log-rank. Se construyeron modelos de supervivencia univariados y multivariados utilizando análisis de regresión de Cox. Se consideró significativo un valor de p menor a 0,05. **Resultados.** Entre nuestra cohorte de 135 pacientes con CM, el 51,1% (69 casos) tenían MA. La mediana de edad fue de 68 años, con predominio de mujeres (55%), y el 88,4% tuvo el sitio plantar como sitio primario. La tasa de supervivencia global a 5 años (SG) fue del 54,3%. En el análisis univariado, el nivel de Clark III/IV, la anaplasia, la invasión linfocítica, el estadio III-IV y el NLR estuvieron asociados con pobre pronóstico. En el análisis multivariado, un NLR >3,5 (HR 3,9, IC del 95% 1,5-10,3, p=0,005) y un nivel de Clark III-IV (HR 3,5, IC del 95% 1,6-7,8, p=0,002) estuvieron asociados con una menor SG. **Conclusión.** El NLR emerge como un factor pronóstico independiente para la SG entre los pacientes peruanos con MA en una institución para manejo del cáncer.

Palabras clave
Melanoma cutáneo maligno; factor pronóstico; sobrevida (fuente: DeCS-BIREME).

INTRODUCTION

Acral melanoma (AM) is a cutaneous melanoma (CM) subtype characterized by location on acral sites, as first described by Reed[1]. While the genesis of non-acral CM is associated to intermittent sun exposure, the etiology of AM remains defined insufficiently[2]. Some studies have included traumatic injury, ultraviolet light exposure and chemical exposure as potential risk factors[3-6]. Incidence rates of AM vary widely among different populations. In Caucasians, AM is a rare neoplasia, accounting for 1%–7% of all CM[7]. Conversely, in Asian and Latin American countries, AM is the most frequent subtype of CM, with Peru having one of the highest reported incidences, ranging between 35% and 61.2%[8,9].

AM is characterized by a poor prognosis, often attributed to diagnosis, and/or its biologically aggressive nature[10,11]. This heightened aggressiveness may stem from its distinct biology compared to non-acral CM, which typically exhibits a high mutational load, thereby enhancing its response to immunotherapy[2]. In contrast, AM typically presents a lower mutational burden, with a low percentage of BRAF and NRAS mutations, and the presence of other proto-oncogenes such as NF1, KIT, MAP2H2 or TERT mutations [12], rendering it less responsive to immunotherapy with checkpoint inhibitors[12,13].

The most clinically relevant prognostic factors for CM include the tumor thickness, safe margins, sentinel lymph node biopsy, and ulceration[2]. While prognostic factors for the AM subtype have not been clearly defined, it has been reported that relevant clinical prognostic factors include tumor thickness and clinical stage[10]. Recently, new prognostic parameters such as the neutrophil-to-lymphocyte ratio (NLR)[14,15], lymphocyte -to-monocyte ratio (LMR)[16], and platelets-to-lymphocyte ratio (PLR)[17] have emerged, which have been widely used in different types of neoplasms. There is currently a need for novel and efficient prognostic biomarkers in CM, especially in AM, which has been less studied in our population. Therefore, our aim was to evaluate whether NLR, LMR, and PLR serve as prognostic biomarkers in AM, the most frequent subtype of CM in Peru.

MATERIALS AND METHODS

The present study utilized an analytical, retrospective observational and cross-sectional design. The study population consisted of patients diagnosed with AM at our institution between 2010 and 2015. Inclusion criteria comprised a histopathological diagnosis of AM, patients aged 18 years or older, availability of complete clinical information and follow-up data. Exclusion criteria were the presence of a second neoplasm and incomplete clinical information.

Study variables

At the time of initial diagnosis of cutaneous melanoma (CM), baseline clinical, laboratory and pathological features were abstracted. We gathered data on the following clinical parameters: age, sex and primary site of CM; laboratory measures including Neutrophil-to-Lymphocyte Ratio (NLR), Lymphocyte-to-Monocyte Ratio (LMR), and Platelet-to-Lymphocyte Ratio (PLR); and pathological covariates such as Clark level, Breslow thickness, degree of anaplasia, presence of ulceration, microsatellitosis, perineural invasion, lymphatic invasion, vascular invasion, nodal involvement, and clinical stage based on the seventh edition of the American Joint Committee on Cancer (AJCC) staging system. Additionally, information on treatment received, including surgery, adjuvant therapy, and first-line treatment, was collected.

Ethical considerations

This project did not involve direct contact or intervention with patients, as it was an observational study conducted through chart reviews. The confidentiality of the obtained information was strictly maintained. Approval for this project was granted by both the Protocol Review Committee and the Ethics Committee at our institution.

Data analysis

Clinical pathological information is presented using descriptive statistics. For the survival analysis, the Kaplan-Meier method was used to generate survival curves, which were compared using the log-rank test. The Cox regression test was used to establish the univariate and multivariate survival models. The results of the Cox model were reported with a hazard ratio (HR) with a 95% confidence interval (CI). The p-value was considered significant when it was less than 0.05. Calculations and graphs were obtained with the statistical program SPSS, version 22.

RESULTS

In our cohort of 135 patients with cutaneous melanoma, it was found that 51.1% of the patients had ALM (69 cases).

Clinical and laboratory features

There was a significant female sex predominance in our study. The mean age of our cohort was 68 years, with 68% of patients being over 60 years old. Among cases of acral lentiginous melanoma (ALM), the plantar site was the most commonly affected, accounting for 88.4% of cases, followed by the palmar site at 8.6%, and the subungual site at 2.9%. Regarding clinical staging, 28.9% of patients were classified as stage I, 34.8% as stage II, 24.6% as stage III, and 5.8% as stage IV. Elevated NLR (>3.5) was observed

in 17.4% of patients, while a low LMR (<0.2) was present in 1.5% of patients. Furthermore, 27.5% of patients exhibited a high PLR (>170) (see Table 1).

Pathological features

In our study, pathological analysis revealed that 13% of patients had a thickness of 2-4 mm, while 37.7% had a thickness greater than 4 mm. Notably, ulceration was notably absent in most cases, with only 36.2% of incidence. Additionally, a significant proportion of patients did not display perineural infiltration, lymphocyte infiltration, or vascular invasion, with rates of 4.3%, 11.6%, and 4.3%, respectively. Anaplasia was present in 5.8% of cases, while microsatellitosis was observed in 1.4% of cases (see Table 2).

Treatment features

Regarding treatment features, surgery was conducted in 82.6% of patients diagnosed with clinical stage I to III, and complete lymph node dissection (CLND) was performed in

Table 1. Clinical and laboratory features of the entire ALM population

	n	%
Patients	69	
Age		
Median age (range)	68 (16, 89)	
<60	22	31.9
>60	47	68.1
Sex		
Female	38	55.1
Male	31	44.9
Primary site CM		
Plantar	61	88.4
Palmar	6	8.6
Subungual	2	2.9
Stage (7th Edition AJCC)		
I	20	28.9
II	24	34.8
III	17	24.6
IV	4	5.8
Unknown	4	5.8
NLR		
<3.5	50	72.5
>3.5	12	17.4
Unknown	7	10.1
LMR		
>0.2	61	88.4
<0.2	1	1.5
Unknown	7	10.1
PLR		
<170	43	62.3
>170	19	27.5
Unknown	7	10.1

CM: Cutaneous melanoma; NLR: Neutrophil-Lymphocyte Ratio (NLR); LMR: Lymphocyte-to-Monocyte Ratio; PLR: Platelet-to-Lymphocyte Ratio.

Table 2. Baseline pathological features of the ALM population

	n	%
Patients	69	
Clark level		
I	7	10.1
II	14	20.3
III	21	30.4
IV	13	18.8
V	6	8.7
Unknown	8	11.6
Breslow (mm)		
0.01-1	13	18.8
1.01-2	15	21.7
2.01-4	9	13.0
> 4	26	37.7
Unknown	6	8.7
Anaplasia		
No	59	85.5
Yes	4	5.8
Unknown	6	8.7
Ulceration		
No	40	58.0
Yes	25	36.2
Unknown	4	5.8
Microsatellitosis		
No	64	92.8
Yes	1	1.4
Unknown	4	5.8
Perineural invasion		
No	62	89.9
Yes	3	4.3
Unknown	4	5.8
Lymphatic invasion		
No	57	82.6
Yes	8	11,6
Unknown	4	5.8
Vascular invasion		
No	62	89.9
Yes	3	4.3
Unknown	4	5.8

Table 3. Treatment received in the entire ALM population

	n	%
Patients	69	
Surgery		
No	12	17.4
Yes	57	82.6
Complete lymph node dissection (Stage III)	13	
No	3	23.1
Yes	10	76.9
Adjuvant treatment (IIB-IIIC)	31	
No	16	51.6
Yes (Interferon alfa-2b)	10	32.3
Unknown	5	16.1
First line treatment	3	
No (BSC) *	3	100
Yes	0	0

BSC: Best supportive care

76.9% of these cases. Adjuvant treatment with Interferon alfa-2b was administered to 32.3% of patients classified with stage IIB to IIIC. Notably, all patients diagnosed with stage IV received best supportive care, with no systemic treatment initiated due to poor performance status (see Table 3).

Survival outcomes

The OS rate at 5 years was 54.3% (see Figure 1). Univariate analysis indicated that Clark IV-V (HR: 1.8, 95% CI: 1.1-3.2, p=0.016), anaplasia (HR: 3.0, 95% CI: 1.5-5.7, p=0.022), lymphocytic invasion (HR: 2.8, 95% CI: 1.6-5.0, p=0.035),

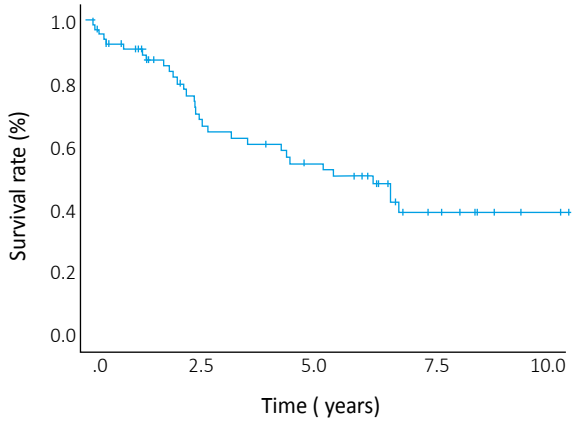

Figure 1. Overall survival in entire AM population

Table 4. Univariate Cox proportional-hazard regression analysis for OS among patients with AM

	Median	5-yr OS (%)	HR	p-value
Overall survival	6.3	54.3	-	-
Age				
<60	NR	52.8	Reference	
>60	5.4	55.1	1.2 (0.7, 2.1)	0.867
Sex				
Female	6.8	63.6	Reference	
Male	3.5	45.1	2.1 (1.3, 3.5)	0.130
Clark level				
I-III	6.7	58.7	Reference	
IV-V	2.5	32.3	1.8 (1.1, 3.2)	0.016
Breslow				
<1	6.3	54.3	Reference	
>1	4.4	42.0	1.2 (0.6, 2.4)	0.644
Anaplasia				
No	5.4	53.6	Reference	
Si	0.4	25.0	3.0 (1.5, 5.7)	0.022
Ulceration				
No	6.3	61.1	Reference	
Si	3.2	41.8	1.8 (1.1, 3.0)	0.453
Lymphatic invasion				
No	6.3	57.4	Reference	
Si	2.2	25.0	2.8 (1.6, 5.0)	0.035
Clinical stage				
I-II	6.7	65.2	Reference	
III-IV	2.5	30.9	2.5 (1.5, 4.1)	0.030
NLR				
<3.5	6.6	57.6	Reference	
>3.5	1.7	31.7	2.1 (1.1, 4.19)	0.002
LMR				
>0.2	6.8	62.5	Reference	
<0.2	4.4	49.6	1.7 (1.0, 3.1)	0.064
PLR				
<170	6.6	58.7	Reference	
>170	4.3	43.6	2.1 (1.2, 3.4)	0.085

NLR: Neutrophil-Lymphocyte Ratio (NLR); LMR: Lymphocyte-to-Monocyte Ratio; PLR: Platelet-to-Lymphocyte Ratio.

advanced clinical stage (HR: 2.5, 95% CI: 1.5-4.1, p=0.030), and NLR > 3.5 (HR: 2.1, 95% CI: 1.1-4.19, p=0.002) were associated with poor prognosis (see Table 4). Multivariate analysis revealed that Clark IV-V (HR: 3.5, 95% CI: 1.6-7.8, p=0.002) and NLR >3.5 were independently associated with lower overall survival (HR: 3.9, 95% CI: 1.5-10.3, p=0.005) (see Table 5 and Figure 2).

Table 5. Multivariate Cox proportional-hazard regression analysis for OS among patients with AM

		p - value	HR	CI 95%	
				Inferior	Superior
Clark level					
I-III	Reference				
IV-V	1.2	0.002	3.5	1.6	7.8
NLR					
<3.5	Reference				
>3.5	1.4	0.005	3.9	1.5	10.3

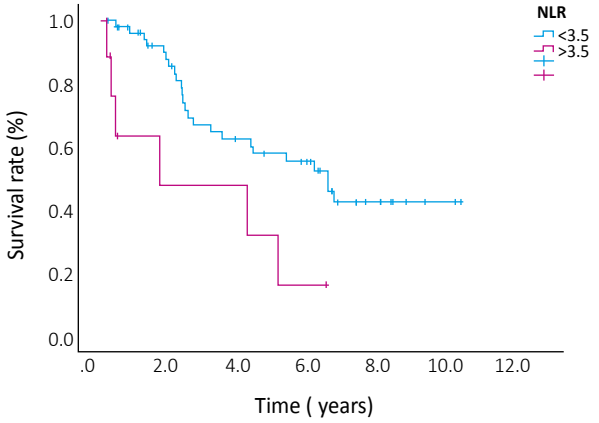

Figure 2. Overall survival in AM stratified by NLR

DISCUSSION

Our study revealed that NLR serves as an independent prognostic biomarker in AM, with an NLR >3.5 being associated with poorer OS. While numerous reports have highlighted this association in CM, studies specific to AM are limited, thus making our study a novel contribution, particularly within the context of Peru and Latin America.

Two metanalysis conducted on CM revealed that an elevated NLR (>3.0) had a significant correlation with shorter OS and progression-free survival (PFS), with the majority of patients being North American and European[18,19]. Zhan et al analyzed twelve studies with 4593 patients with CM and found that an elevated NLR had a significant OS (HR: 1.56, 95% CI: 1.28–1.90, p<0.001) and disease-free survival (DFS)/progression-free survival (PFS) (HR: 1.86; 95% CI: 1.24-2.80; p=0.003); all of these regardless of the clinical stage and the NLR cut-off value[18]. In a more recent metanalysis that included 13 studies from a broader range of countries in Europe, North America and Asia, it was shown that a high NLR predicted poor OS and PFS in patients treated with immunotherapy (HR: 1.71, 95% CI: 1.40-2.10, p<0.001)[20].These finding were consistent regardless of the clinical stage and the NLR cut-off value.

Additionally, some studies reported that a high NLR was associated with treatment failure in patients with advanced CM that received immunotherapy with anti-PD1 or with either BRAF inhibitors alone or combined with MEK inhibitors [21,22]. However, interestingly, a study demonstrated that patients who experienced immune-mediated adverse events and had a high NLR before the second cycle of immunotherapy exhibited higher rates of complete and partial response in advanced melanoma[23]. This data not only suggests that baseline tumoral microenvironment influences the OS prognosis regardless of the type of treatment but also indicates that when combined with immune mediated events, it could potentially reverse the outcomes and predicts higher response rates.

In AM, the significance of the NLR remains understudied, with limited research from few Asian populations investigating its prognostic value[24,25]. Asian studies have identified NLR as a prognostic factor in both early and advanced stages of AM. In the early stage, Yu et al. demonstrated that patients treated with IFNα-2b and an NLR ≥2.35 exhibited poor recurrence-free survival (RFS) and OS[24]. Similarly, in advanced disease, Jung et al. found that a high NLR ≥5 was an independent factor of inferior PFS and OS[25].Additionally, Lee ***et al.***, in a retrospective cohort of 152 patients, including 58 patients (38%) with AM, observed that an NLR >2.1 was associated with worse PFS (median 6.9 vs. 2.4 months, p=0.015) and OS (median not reached vs. 10.4 months, p<0.001)[26].

Interestingly, a recent single Korean study investigated the baseline NLR disparities between AM and non-acral CM, as well as its prognostic significance in patients with AM. The study revealed that the median NLR for AM significantly exceeded that of non-acral CM (2.18 vs. 1.74, p=0.029). In AM, a high NLR (HR: 1.64; 95% CI: 1.02-2.66; p=0.043) was independently associated with poor PFS after adjusting for ulceration, Breslow thickness of ≥2 mm, and nodal invasion[27]. Additionally, a recent Peruvian study demonstrated a strong association between pretreatment NLR ≥ 3 and a higher mortality risk (5-year survival: 22%, and 10-year survival: 14.8%) compared to NLR <3 (5-year survival: 52.7%, and 10-year survival: 41.1%) in all CM[28].

Biomarkers in peripheral blood have become the focus of research in recent years, with their prognostic and predictive value in immunotherapy analyzed across various neoplasms[19,29]. Baseline and post-treatment absolute counts of lymphocytes, eosinophils, neutrophils, and monocytes, as well the NLR, have emerged as promising tools[30].The rationale behind the NLR lies in its ability to measure both the tumor inflammatory response (neutrophilia) and the host immune response (lymphopenia)[31-34]. Neutrophils exhibit a dual role

within the tumor microenvironment, classified into two subtypes: high-density neutrophils (HDN) and low-density neutrophils (LDN). HDN subtype has antitumor activity by directly affecting tumor cells or indirectly by stimulating T-cell mediated immunity. Conversely, the LDN subtype exerts a pro-tumoral activity that favors progression[29]. In cancer, chronic inflammation fosters the accumulation of LDN phenotype, resulting in tumor progression [29]. Additionally, lymphopenia correlates with reduced host immunity and indirectly by the stimulation of suppressor T-cells [34]. These cells contribute to decreased antitumor immune activity, hindering the immune response and inhibiting antitumor immune response. The accumulation of Tregs in cancer has been related to poor outcomes[35,36].

In addition to these findings, we found that our 5-year OS rate was observed to be lower at 54.3% compared to the survival rate among Caucasian population with AM, which stood at 60.5%[37]. In contrast, our results were comparable to those of a Colombian study, which reported an OS of 54%[38], but slightly higher when compared to the Asian population, where OS rates ranged between 41.5 and 49.3%[3,6]. Furthermore, a recent larger study conducted in Peru revealed that the AM subtype had a significantly reduced OS when compared with Non-acral CM subtype (34.7 vs. 59.4% at 5 years, p=0.001)[39].

Our study had several limitations. Firstly, it was a retrospective study with a small sample size of AM patients compared with other series, which limited the power of the study. Additionally, it was conducted at a single cancer center. Therefore, the generalizability of our findings may be limited. However, our institution receives an important part of CM cases in Peru, suggesting that our findings could reasonably apply to a broader segment of this insured population. Nonetheless, there may also be a selection bias inherent in our study design.

Conversely, this study provides valuable insights by raising awareness of the influence of the tumor microenvironment through NLR as a significant prognostic biomarker in both early and advanced scenarios in AM.

This contribution is particularly noteworthy given the scarcity of such evidence previously reported in Peru and Latin America. More importantly, it could guide clinicians in determining treatment strategies based on NLR status at baseline in AM, thereby paving the way for a deeper understanding of its tumoral microenvironment.

In conclusion, following these findings, our study confirms the role of NLR as a prognostic biomarker in AM. This is remarkable in the sense that it could have the potential not only to predict prognosis but also predict treatment responses with immunotherapy and targeted therapy. Nevertheless, larger prospective studies are needed to validate and confirm our findings. Collaborative endeavors among Latin American countries are crucial to gaining a more comprehensive understanding of the unique biological behavior of AM, including its ethnic variations, treatment modalities and survival outcomes.

REFERENCES

1. Krausz K. Pathology of Melanocytic Disorders. 2nd Edition. London: CRC Press; 2012.
2. Schadendorf D, van Akkooi ACJ, Berking C, Griewank KG, Gutzmer R, Hauschild A, *et al.* Lancet. 2018;392(10151):971-984. doi: 10.1016/S0140-6736(18)31559-9. Erratum in: Lancet. 2019;393(10173):746.
3. Jung HJ, Kweon SS, Lee JB, Lee SC, Yun SJ. A clinicopathologic analysis of 177 acral melanomas in Koreans: relevance of spreading pattern and physical stress. JAMA Dermatol. 2013;149(11):1281-8. doi: 10.1001/jamadermatol.2013.5853.
4. Piliang MP. Acral Lentiginous Melanoma. Surg. Pathol. Clins. 2009;2(3):535-41.
5. Phan A, Touzet S, Dalle S, Ronger-Savlé S, Balme B, Thomas L. Acral lentiginous melanoma: a clinicoprognostic study of 126 cases. Br J Dermatol. 2006;155(3):561-9. doi: 10.1111/j.1365-2133.2006.07368.x.
6. Lv J, Dai B, Kong Y, Shen X, Kong J. Acral Melanoma in Chinese: A Clinicopathological and Prognostic Study of 142 cases. Sci Rep. 2016;6:31432. doi: 10.1038/srep31432.
7. Bradford PT, Goldstein AM, McMaster ML, Tucker MA. Acral lentiginous melanoma: incidence and survival patterns in the United States, 1986-2005. Arch Dermatol. 2009;145(4):427-34. doi: 10.1001/archdermatol.2008.609.
8. Coras N, Morales D, Yabar A, Beltran BE. Prognosis of melanoma in Peru: An analysis of 410 cases. ASCO Meet Abstr [Internet]. 2013;31(15_suppl):e20023. Available from: http://meeting.ascopubs.org/cgi/content/abstract/31/15_suppl/e20023
9. Lozano-Espinoza N, Ramos W, Galarza C, Cerrillo G, Tello M, Gutierrez EL. Melanoma cutáneo y mucoso: epidemiología, características clínicas y metástasis a distancia en un hospital de Lima-Perú. Periodo 1996-2007. Dermatol Peru. 2009;19(4):314-21.
10. Asgari MM, Shen L, Sokil MM, Yeh I, Jorgenson E. Prognostic factors and survival in acral lentiginous melanoma. Br J Dermatol. 2017;177(2):428-435. doi: 10.1111/bjd.15600.
11. Bradford PT, Goldstein AM, McMaster ML, Tucker MA. Acral lentiginous melanoma: Incidence and survival patterns in the United States, 1986-2005. Arch Dermatol. 2009;145(4):427–34.
12. Darmawan CC, Jo G, Montenegro SE, Kwak Y, Cheol L, Cho KH, *et al.* Early detection of acral melanoma: A review of clinical, dermoscopic, histopathologic, and molecular characteristics. J Am Acad Dermatol. 2019;81(3):805-12. doi: 10.1016/j.jaad.2019.01.081.
13. Klemen ND, Wang M, Rubinstein JC, Olino K, Clune J, Ariyan S, *et al.* Survival after checkpoint inhibitors for metastatic acral, mucosal and uveal melanoma. J Immunother Cancer. 2020;8(1):e000341. doi: 10.1136/jitc-2019-000341.

14. Annibali O, Hohaus S, Marchesi F, Cantonetti M, Di Rocco A, Tomarchio V, *et al.* The neutrophil/lymphocyte ratio ≥3.5 is a prognostic marker in diffuse large B-cell lymphoma: a retrospective analysis from the database of the Italian regional network 'Rete Ematologica del Lazio per i Linfomi' (RELLI). Leuk Lymphoma. 2019;60(14):3386-3394. doi: 10.1080/10428194.2019.1633628.
15. Chen J, Deng Q, Pan Y, He B, Ying H, Sun H, *et al.* Prognostic value of neutrophil-to-lymphocyte ratio in breast cancer. FEBS Open Bio. 2015;5:502-7. doi: 10.1016/j.fob.2015.05.003.
16. Wan L, Wu C, Luo S, Xie X. Prognostic Value of Lymphocyte-to-Monocyte Ratio (LMR) in Cancer Patients Undergoing Immune Checkpoint Inhibitors. Dis Markers. 2022;2022:3610038. doi: 10.1155/2022/3610038.
17. Xu H, He A, Liu A, Tong W, Cao D. Evaluation of the prognostic role of platelet-lymphocyte ratio in cancer patients treated with immune checkpoint inhibitors: A systematic review and meta-analysis. Int Immunopharmacol. 2019;77:105957. doi: 10.1016/j.intimp.2019.105957.
18. Zhan H, Ma JY, Jian QC. Prognostic significance of pretreatment neutrophil-to-lymphocyte ratio in melanoma patients: A meta-analysis. Clin Chim Acta. 2018;484:136-140. doi: 10.1016/j.cca.2018.05.055.
19. Ding Y, Zhang S, Qiao J. Prognostic value of neutrophil-to-lymphocyte ratio in melanoma: Evidence from a PRISMA-compliant meta-analysis. Medicine (Baltimore). 2018;97(30):e11446. doi: 10.1097/MD.0000000000011446.
20. Zhang Y, Liu B, Kotenko S, Li W. Prognostic value of neutrophil-lymphocyte ratio and lactate dehydrogenase in melanoma patients treated with immune checkpoint inhibitors: A systematic review and meta-analysis. Medicine (Baltimore). 2022;101(32):e29536. doi: 10.1097/MD.0000000000029536.
21. Cocorocchio E, Martinoli C, Gandini S, Pala L, Conforti F, Stucchi S, *et al.* Baseline neutrophil-to-lymphocyte ratio (NLR) is associated with outcome of patients treated with BRAF inhibitors. Clin Transl Oncol. 2020;22(10):1818-1824. doi: 10.1007/s12094-020-02320-y.
22. Bartlett EK, Flynn JR, Panageas KS, Ferraro RA, Jessica JM, Postow MA, *et al.* High neutrophil-to-lymphocyte ratio (NLR) is associated with treatment failure and death in patients who have melanoma treated with PD-1 inhibitor monotherapy. Cancer. 2020;126(1):76-85. doi: 10.1002/cncr.32506.
23. Mesti T, Grašič Kuhar C, Ocvirk J. Biomarkers for Outcome in Metastatic Melanoma in First Line Treatment with Immune Checkpoint Inhibitors. Biomedicines. 2023;11(3):749. doi: 10.3390/biomedicines11030749
24. Yu J, Wu X, Yu H, Li S, Mao LL, Chi Z, *et al.* Systemic Immune-Inflammation Index and Circulating T-Cell Immune Index Predict Outcomes in High-Risk Acral Melanoma Patients Treated with High-Dose Interferon. Transl Oncol. 2017;10(5):719-725. doi: 10.1016/j.tranon.2017.06.004.
25. Jung M, Lee J, Kim TM, Lee DH, Kang JH, Oh SY, *et al.* Ipilimumab Real-World Efficacy and Safety in Korean Melanoma Patients from the Korean Named-Patient Program Cohort. Cancer Res Treat. 2017;49(1):44-53. doi: 10.4143/crt.2016.024.
26. Lee J, Lee SJ, Kim K, Kim ST, Jang KT, Lee J. Comprehensive molecular and clinical characterization of Asian melanoma patients treated with anti-PD-1 antibody. BMC Cancer. 2019;19(1):805. doi: 10.1186/s12885-019-6030-5.
27. Choi ME, Jung JM, Kim DH, Won CH, Chang SE, Lee MW, *et al.* Baseline Serum neutrophil-to-lymphocyte ratio in acral melanoma compared with nonacral melanoma and its prognostic significance. J Am Acad Dermatol. 2024;90(5):977-985. doi: 10.1016/j.jaad.2023.12.064.
28. Pinto-Paz ME, Cotrina-Concha JM, Benites-Zapata VA. Mortality in cutaneous malignant melanoma and its association with Neutrophil-to-Lymphocyte ratio. Cancer Treat Res Commun. 2021;29:100464. doi: 10.1016/j.ctarc.2021.100464.
29. Sacdalan DB, Lucero JA, Sacdalan DL. Prognostic utility of baseline neutrophil-to-lymphocyte ratio in patients receiving immune checkpoint inhibitors: a review and meta-analysis. Onco Targets Ther. 2018;11:955-965. doi: 10.2147/OTT.S153290.
30. Kitano S, Nakayama T, Yamashita M. Biomarkers for Immune Checkpoint Inhibitors in Melanoma. Front Oncol. 2018;8:270. doi: 10.3389/fonc.2018.00270.
31. Wade RG, Robinson A V., Lo MCI, Keeble C, Marples M, Dewar DJ, *et al*. Baseline Neutrophil-Lymphocyte and Platelet-Lymphocyte Ratios as Biomarkers of Survival in Cutaneous Melanoma: A Multicenter Cohort Study. Ann Surg Oncol. 2018;25(11):3341-3349. doi: 10.1245/s10434-018-6660-x.
32. Davis JL, Langan RC, Panageas KS, Zheng J, Postow MA, Brady MS, *et al.* Elevated Blood Neutrophil-to-Lymphocyte Ratio: A Readily Available Biomarker Associated with Death due to Disease in High Risk Nonmetastatic Melanoma. Ann Surg Oncol. ' 2017;24(7):1989-1996. doi: 10.1245/s10434-017-5836-0.
33. Ferrucci PF, Gandini S, Battaglia A, Alfieri S, Di Giacomo AM, Giannarelli D, *et al*. Baseline neutrophil-to-lymphocyte ratio is associated with outcome of ipilimumab-treated metastatic melanoma patients. Br J Cancer. 2015;112(12):1904-10. doi: 10.1038/bjc.2015.180.
34. Cassidy MR, Wolchok RE, Zheng J, Panageas KS, Wolchok JD, Coit D, *et al.* Neutrophil to Lymphocyte Ratio is Associated With Outcome During Ipilimumab Treatment. EBioMedicine. 2017;18:56-61. doi: 10.1016/j.ebiom.2017.03.029.
35. Aranda F, Vacchelli E, Obrist F, Eggermont A, Galon J, Sautès-Fridman C, *et al.* Trial Watch: Toll-like receptor agonists in oncological indications. Oncoimmunology. 2014;3:e29179. doi: 10.4161/onci.29179.
36. Ladányi A. Prognostic and predictive significance of immune cells infiltrating cutaneous melanoma. Pigment Cell Melanoma Res. 2015;28(5):490-500. doi: 10.1111/pcmr.12371.
37. Csányi I, Houshmand N, Szűcs M, Ócsai H, Kemény L, Oláh J, *et al*. Acral lentiginous melanoma: a single-centre retrospective review of four decades in East-Central Europe. J Eur Acad Dermatol Venereol. 2020;34(9):2004-2010. doi: 10.1111/jdv.16227.
38. Duarte CA, Flórez JP, López HG, Meneses MX, de Vries E. Survival of acral lentiginous melanoma in the National Cancer Institute of Colombia. J Eur Acad Dermatol Venereol. 2017;31(3):438-442. doi: 10.1111/jdv.13913.
39. Castaneda CA, Torres-Cabala C, Castillo M, Villegas V, Casavilca S, Cano L, *et al.* Tumor infiltrating lymphocytes in acral lentiginous melanoma: a study of a large cohort of cases from Latin America. Clin Transl Oncol. 2017;19(12):1478-1488. doi: 10.1007/s12094-017-1685-3.