

Relationship of the prognostic nutritional index with complications and mortality in patients with gastric cancer who underwent gastrectomy in a tertiary hospital in Bogotá, Colombia

Relación del índice nutricional pronostico con complicaciones y mortalidad en los pacientes con cáncer gástrico sometidos a gastrectomía en un hospital de tercer nivel de Bogotá, Colombia

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Abstract

Introduction. Gastric cancer is the fifth most frequently diagnosed cancer and the third cause of cancer death in the world. In surgical treatment, current evidence supports preoperative measures and prognostic indices to improve survival. The prognostic nutritional index, which unites the values of circulating lymphocytes in peripheral blood with those of serum albumin, has presented characteristics of being a nutritional and immunological marker with predictive value on complications and mortality. The objective of this study was to determine the relationship between the prognostic nutritional index with complications and mortality in patients with gastric cancer undergoing gastrectomy.

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Methods. A descriptive, cross-sectional, observational study with an analytical component was carried out by a retrospective review of medical records.

Results. A total of 113 patients who underwent total or subtotal gastrectomy were analyzed. An association was found between mortality and the prognostic nutritional index and mortality; all patients who died had an index \leq 46. An inverse association was also found between the value of the index and the presentation of postoperative complications, such as sepsis, peritonitis, anastomotic leak, and bleeding.

Discussion. Similar to our analysis, several studies suggest that a low prognostic nutritional index could have a predictive value on the frequency of complications and overall survival in patients with gastric cancer undergoing surgery.

Conclusion. The prognostic nutritional index is associated with mortality and postoperative complications in patients undergoing gastrectomy for gastric cancer.

Keywords: stomach neoplasms; nutrition assessment; gastrectomy; postoperative complications; mortality; prognosis.

Resumen

Introducción. El cáncer gástrico es el quinto cáncer diagnosticado con mayor frecuencia y la tercera causa de muerte por cáncer en el mundo. En el tratamiento quirúrgico, la evidencia actual apoya las medidas preoperatorias e índices pronósticos para mejorar la supervivencia. El índice nutricional pronóstico, que une los valores de los linfocitos circulantes en sangre periférica con los de la albúmina sérica, ha presentado características de ser un marcador nutricional e inmunológico con valor predictivo sobre complicaciones y mortalidad. El objetivo de este estudio fue determinar la relación entre el índice nutricional pronostico con las complicaciones y mortalidad en pacientes con cáncer gástrico sometidos a gastrectomía.

Métodos. Se llevó a cabo un estudio observacional descriptivo, de corte transversal, con componente analítico, mediante la revisión retrospectiva de las historias clínicas.

Resultados. Se analizaron 113 pacientes sometidos a gastrectomía total o subtotal. Se encontró asociación entre el índice nutricional pronóstico y la mortalidad; todos los pacientes que murieron tenían un índice menor o igual a 46. También se encontró asociación inversa entre el valor del índice y la presentación de complicaciones posoperatorias, como sepsis, peritonitis, fuga de la anastomosis y sangrado.

Discusión. Similar a nuestro análisis, varios estudios plantean que un índice nutricional pronostico bajo podría tener un valor predictivo sobre la frecuencia de complicaciones y supervivencia global en pacientes con cáncer gástrico llevados a cirugía.

Conclusión. El índice nutricional pronostico se asocia con la mortalidad y complicaciones posoperatorias en pacientes sometidos a gastrectomía por cáncer gástrico.

Palabras clave: neoplasias gástricas; evaluación nutricional; gastrectomía; complicaciones posoperatorias; mortalidad; pronóstico.

Introduction

In recent years, the incidence of gastric cancer (GC) has decreased, however, and despite improvements in surgical and oncological treatments, mortality

remains very high ¹⁻⁵. There are differences in the incidence of GC according to its geographical distribution: the areas with the highest incidence are Asia (mainly Japan), western South America, and

eastern Europe. There is also notable variability in the natural history of the disease among patients, especially in survival. The survival rate 5 years after diagnosis is 20-30% in Western countries $^{6-12}$. Colombia is among the countries with the highest incidence rate (17.4 to 48.2 per 100,000 inhabitants), and here GC is the leading cause of death from cancer in men and the third in women, so this tumor represents a real public health problem.

The treatment of GC is fundamentally based on radical surgery ¹³. To achieve this, first is necessary to assess whether the patient is fit to be operated on and whether the tumor is resectable. Resectability, at the time of diagnosis, can only be presumed by clinical TNM classification, whose accuracy is limited and tends to understaging in at least 20% of cases, so it is a criterion that is only confirmed after surgery. Therefore, it is necessary to have other predictive parameters, independent of the clinical TNM, and that can be determined at the time of tumor diagnosis, such as immunity, inflammation and nutrition ^{12,14}.

The patient with GC presents many symptoms that ultimately lead to a state of malnutrition ¹⁴; rapid growth of malignant tumor cells causes consumption of large amount of nutrients, lack of synthesis of nutrients, necrosis and production of toxic substances, leading to metabolic disorders, therefore, most patients have different degrees of malnutrition at the time of treatment ^{15,16}. Malnutrition can delay treatment, which can lead to disease progression. Furthermore, malnutrition leads to decreased T-cell function and deterioration of the intestinal environment, further aggravating the progression of tumor-associated inflammation ¹⁷. In 1863, Virchow first proposed the association between inflammation and cancer, and since then, the role of inflammation in tumor genesis, tumor development, and metastasis has been continuously explored ¹⁸. Mantovani et al ¹⁹ showed that tumor-associated inflammation is characterized by the release of inflammatory factors, which further favor tumor development, invasion, and metastasis ²⁰⁻²³.

Malnutrition causes changes in body weight, muscle mass, serum albumin levels, lymphocyte

count, number of helper T cells, interleukin levels, and blastogenic response. The cumulative consequences of these alterations adversely affect the prognosis of cancer patients, through increased postoperative complications, attributed to tissue vulnerability, impaired wound healing, susceptibility to infection, and accelerated tumor progression ²⁴.

The Prognostic Nutritional Index (PNI) was first established by Japanese scholars and was originally used to assess preoperative nutritional status, surgical risk, and postoperative complications in surgical patients ¹. The Onodera Index ¹⁰, also called INP, combines the values of circulating lymphocytes and those of serum albumin. It has been proposed as a predictor of prognosis for patients with gastrointestinal neoplasms ²⁵, colorectal cancer ²⁶, liver cancer ²⁷, and pancreatic cancer ²⁸; reflecting immunocompetence and nutritional status more accurately ²⁴.

The nutritional prognostic index of Onodera (Onodera's nutritional prognostic index, OPNI) is calculated with the following formula ²⁹:

OPNI = 10 X Albumin (g/dl) + 0.005 X circulating lymphocytes / mm³

Ouyang et al ⁵ showed that preoperative serum albumin levels are associated with prognosis in patients with GC. The immune response of lymphocytes to tumor has been gradually applied to prognostic evaluation in cancer patients in recent years ²⁵. A recent study shows that preoperative PNI is superior to traditional inflammatory markers for prognostic evaluation after gastrectomy and that it is an independent risk factor for mortality after gastrectomy ³⁰. This can be explained because the preoperative PNI reflects the nutritional status and the inflammatory response of the body.

The objective of this study was to determine the relationship between preoperative PNI with complications, reinterventions and mortality in patients with gastric cancer undergoing gastrectomy.

Methods

A descriptive, cross-sectional, observational study with an analytical component was carried out, including patients with gastric cancer who underwent total or subtotal gastrectomy, between 2014 and 2019, at the Hospital Universitario Clínica San Rafael in the city of Bogotá, Colombia. Patients who did not have a complete clinical history, without the possibility of completing the data, who did not agree to be interviewed by telephone when the data collection warranted it, those who had a synchronous or metachronous neoplasia, with some infectious or autoimmune disease or with chronic steroid use were excluded.

Probabilistic sampling was performed with a confidence of 95%, a power of 90%, an error of 5%. The sample size was calculated in 113 patients, who were selected according to the random sequence. The source of information and unit of analysis was the clinical history. For the collection and consignment of the study variables, a data collection form, a database and a telephone booklet were designed.

An exploratory data analysis was performed to describe the sample, where discrete variables were shown as percentages and sample size, and continuous variables were presented as mean, median, standard error, and sample size. The Shapiro-Wilk test was applied to determine if the continuous variables had a normal distribution; Chi-square test or Fisher's exact test, to determine the relationship between the discrete variables and the INP, with their OR and confidence intervals; Spearman-Pearson correlation was calculated to determine the relationship of continuous variables with PNI, multiple logistic regression, with which variables related to complications and Kapplan-Meyer curve were found among patients who presented mortality with the test of Wilcoxon.

Results

One-hundred-thirteen patients who underwent total or subtotal gastrectomy for gastric cancer were included, whose clinical and pathological characteristics are presented in Table 1. The mean age of the study cohort was 64 years and most of the patients were male; 38.9% of the patients had cardiovascular comorbidities. Total gastrectomy was performed in 63.7% of the patients and half received neoadjuvant chemotherapy. Some type of complication occurred in 39.8% of the patients, with the need for additional surgery in 25% of the cases.

The nutritional characteristics of the patients are shown in Table 2 and the laboratory tests in Table 3. Most of the patients (61.1%) had a level of nutrition classified as healthy weight. Patients lost an average of almost eight kilograms of weight in the last 6 months, and up to half of the patients lost seven kilograms of weight. Most of the patients (67.3%) received parenteral nutrition and almost half of the patients received enteral nutrition. Regarding the PNI, up to half of the patients had a score of 38.4.

Half of the patients presented tumors located in the gastric antrum or with a Borrmann III macroscopic classification, that is, an infiltrating and

Table 1. Clinicopathological characteristics of patients undergoing gastrectomy (n=113 patients).

Frequency (%)
64 (1.3)
62 (54.9)
51 (45.1)
44 (38.9)
17 (15)
37 (32.7)
3 (2.7)
72 (63.7)
41 (36.3)
58 (51.3)
45 (39.8)
12 (10.6)
10 (8.8)
23 (20.4)
31 (27.4)
3 (2.7)
2 (1.8)
11 (9.7)
29 (25.7)

Variable	Frequency (%)
Weight (kg), mean (standard deviation)	59.2 (1.3)
Height (m), mean (standard deviation)	1.6 (0)
Body mass index (kg/m2), mean (standard deviation)	23 (0.5)
Nutrition status	
Very severe thinness	2 (1.8)
Severe thinness	2 (1.8)
Thinness	14 (12.4)
Healthy weight	69 (61.1)
Overweight	17 (15)
Overweight grade 1	5 (4.4)
Overweight grade 2	3 (2.7)
Overweight grade 3	0 (0)
Overweight grade 4	1 (0.9)
Weight loss in the last 6 months, mean (standard deviation)	7.9 (0.6)
Parenteral nutrition	76 (67.3)
Enteral nutrition	55 (48.7)
Predictive Nutritional Index (PNI), mean (standard deviation)	37.7 (0.7)

 Table 2. Nutritional characteristics of patients undergoing gastrectomy (n=113 patients).

 Table 3. Paraclinical characteristics of patients undergoing gastrectomy (n=113 patients).

Variable	Average	SD	Median
White blood cell count (cell/mm3)	8451.5	551.3	7400.0
Neutrophil ratio (%)	70.1	1.1	71.0
Lymphocyte ratio (%)	12.2	0.7	11.0
Absolute lymphocyte count (cell/mm3)	949.0	64.0	835.9
Hemoglobin (gr/l)	11.5	0.3	11.8
Platelets (cel/mm3)	317.293.2	9870.3	304.000.0
Albumin level (gr/dl)	3.3	0.1	3.3
Prealbumin level (mg/dl)	15.6	0.7	14.2
Total proteins (gr/dl)	5.6	0.1	5.4
Total cholesterol level (mg/dl)	133.0	3.7	137.0
Triglycerides (mg/dl)	103.5	3.2	103.0
Transferrin (mg/dl)	245.6	6.9	253.0
Ferritin (ng/ml)	34.8	2.9	31.5

ulcerated tumor (Table 4). The most frequent clinical stages were IIA, IB and IIB. Regarding tumor histology, the highest proportion were well-differentiated and intestinal adenocarcinomas in the Lauren classification.

Patients had an average hospital stay of 9 days before surgery, and complications were identified

Table 4. Oncological clinical characteristics of patients undergoing gastrectomy (n=113 patients).

Variable	Frequency (%)
Macroscopic extent of the tumor	
Borrmann V	5 (4.4)
Borrmann IV	29 (25.7)
Borrmann III	58 (51.3)
Borrmann II	21 (18.6)
Clinical stage of the tumor	
IA	10 (8.8)
IB	18 (15.9)
IIA	40 (35.4)
IIB	13 (11.5)
IIIA	10 (8.8)
IIIB	4 (3.5)
IIIC	4 (3.5)
IV	14 (12.4)
Tumor histology	
World Health Organization (WHO) Classification	
Well-differentiated adenocarcinoma	41 (36.3)
Signet ring cell carcinoma	32 (28.3)
Poorly differentiated adenocarcinoma	20 (17.7)
Poorly cohesive carcinoma	5 (4.4)
Mucinous adenocarcinoma	5 (4.4)
Others	10 (8.8)
Lauren's Classification	
Intestinal	66 (58.4)
Difuse	27 (23.9)
Undetermined	20 (17.7)
Tumor location	
Cardias	9 (8)
Fundus	8 (7.1)
Body	30 (26.5)
Anthro	55 (48.7)
Pylorus	11 (9.7)

on average 2 days after surgery; 61% required an Intensive Care Unit, with an average stay of almost four days.

The clinical, pathological and nutritional factors that were significant in the multivariate analysis were included in Table 6. A significant association was found between PNI and mortality; all the patients who died had a PNI less than or equal to 46.03, and of the total number of patients with a PNI less than or equal to 46.03, 24.2% died.

The analysis and correlation of the PNI with the continuous variables is outlined in Table 7. A statistically significant positive relationship

Table 5.	Characteristics	related to	hospitalization.
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Variable	Value
Days of hospital stay prior to surgery, mean (± standard deviation)	9.1 (± 0.7)
Post-operative days at the time complications were identified, mean (± standard deviation)	2.1 (± 0.3)
Management in the Intensive Care Unit (ICU)	59 (61.1%)
Management in the Special Care Unit (UCE)	22 (19.5%)
Days of ICU stay, mean (± standard deviation)	3.9 (± 0.6)
Days of stay in CSU, mean (± standard deviation)	1.1 (± 0.3)
Days of hospital stay, mean (± standard deviation)	20.6 (± 1.3)

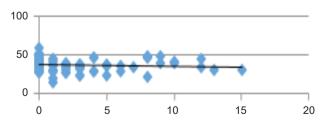
was identified between the PNI with the value of hemoglobin, prealbumin, total proteins, total cholesterol and transferrin, as well as an inverse relationship between the PNI and the post-surgical days at the time of identifying the complications (Figure 1). For the other variables there was no statistically significant difference, but established trends were observed.

Figure 2 shows the survival curve, calculated independently of the PNI value, where it is observed that the patients had an 80% probability of surviving up to two years.

In logistic regression, an inverse association was found between the PNI value and the presentation of postoperative complications such as sepsis, peritonitis, anastomosis leak, bleeding, and mortality, as well as the level of hemoglobin and prealbumin in the exploratory analysis (Table 8).

Table 6. Relationship between the prognostic nutritional index (PNI) and mortality in patients undergoing gastrectomy (n=113 patients).

Mortality	/	PNI ≤ 46,03	PNI > 46,03	Total	р
	n	24	0	24	
Death	% Mortality	100.0	0.0	100.0	
Death	PNI less than 46.03	24.2%	0.0%	21.2%	
	% of the total	21.2	0.0	21.2	
Alive	n	75	14	89	_
	% Mortality	84.3	15.7	100.0	0.038
	PNI less than 46.03	75.8%	100.0%	78.8%	
	% of the total	66.4	12.4%	78.8%	



Source: Real Statitistics V7.6 Jan 2121, R V

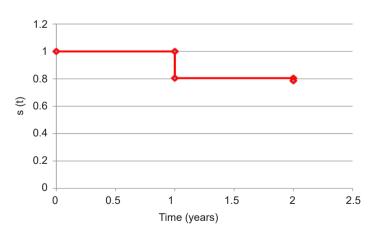
Figure 1. Post-surgical days at the time of identifying complications.

/ariable	Sw p *	Rho (ρ) **	р
Age (years)	0.008	-0.06	0.518
Neight (kg)	0.001	0.10	0.271
Height (cm)	0.257	-0.04	0.722
Body Mass Index	0.000	0.14	0.137
Neight loss in the last 6 months	0.000	0.02	0.853
Hemoglobin (gr/l)	0.528	0.25	0.012
White blood cell count (cell/mm3)	0.000	0.06	0.504
Neutrophil count (%)	0.027	-0.12	0.203
Platelet count (cell/mm3)	0.007	0.09	0.330
Prealbumin level (mg/dl)	0.053	0.28	0.000
Total proteins (gr/dl)	0.015	0.48	0.000
Total cholesterol (mg/dl)	0.013	0.22	0.017
Triglycerides (md/dl)	0.033	-0.07	0.456
⁼ erritin (ng/ml)	0.000	0.00	0.970
Transferrin (mg/dl)	0.080	0.18	0.020
Parenteral nutrition (days)	0.000	-0.16	0.093
Enteral nutrition (days)	0.000	-0.09	0.356
Days of hospital stay prior to surgery	0.000	-0.17	0.075
Post-surgical days at the time of identifying complications	0.000	-0.27	0.004
ntensive Care Unit stay (days)	0.000	-0.03	0.721
Special Care Unit stay (days)	0.000	-0.12	0.210
Hospital stay (days)	0.000	0.00	0.974

Table 7. Correlation between the prognostic nutritional index (PNI) and continuous variables in patients undergoing gastrectomy (n=113 patients).

* Shapiro-Wilk test, less than 0.05, non-normal distribution

** Spearman correlation



Source: Real Statitistics V7.6 Jan 2121, R V

Figure 2. Survival curve.

Discussion

Some studies ⁴⁻⁶ have indicated that the depth of invasion and the presence or absence of lymph node metastases are the most important prognostic factors in gastric cancer. Others focus their hypotheses on tumor markers that come mainly from the cancer itself as a prognostic factor ⁷.

Prognostic value has recently been given to nutritional markers as a predictor of results in patients with GC undergoing total gastrectomy. There are several tools to assess nutritional status, among which the simplest and most convenient have traditionally been the Body Mass Index (BMI) and serum proteins, such as albumin and prealbumin ⁴⁻⁶.

Ouyang et al ⁵ demonstrated that preoperative serum albumin levels, a parameter included in the PNI, is associated with prognosis in patients with GC. Albumin has been commonly used to determine the nutritional status of patients and a study revealed that serum albumin is an important factor in predicting prognosis in patients with GC, however, the level of serum albumin is an unreliable tool since it can be influenced by systemic inflammatory reactions ²⁴. As in these studies, in ours it had no predictive value as an independent variable.

Mantovani et al ¹⁹ demonstrated that tumor-associated inflammation is characterized by the proliferation of inflammatory cells and the production and release of inflammatory factors in tumor tissues. Inflammatory cells and immune cells, such as neutrophils, lymphocytes, and monocytes, in the peripheral blood of patients with tumor-associated inflammation are considered important factors leading to tumor development, invasion, and metastasis. Like our study, other studies have shown that nutritional indices and inflammatory parameters play an important role in determining the prognosis of cancer patients ³¹⁻³³.

Complication		PNI > 46,03	Hemoglobin < 10 g/dl	Prealbumin < 8 mg/dl
	Coeff b	-0.18	-1.56	-0.13
Sepsis	Valor p	0.04	0.005	0.24
	OR (IC _{95%})	0.84 (0.67 - 1.04)	4.77 (1.61 - 14.1)	0.87 (0.7 - 1.09)
	Coeff b	-1.38	-2.31	-0.5
Peritonitis	Valor p	0.001	0.04	0.1
	OR (IC _{95%})	0.9 (0.5 - 0.97)	10.05 (8 - 20)	1.65 (1.4 - 1.9)
	Coeff b	-0.12	-0.04	-0.02
Anastomotic leak	Valor p	0.01	0.08	0.02
	OR (IC _{95%})	0.88 (0.77 - 1.01)	0.96 (0.7 - 1.31)	1.98 (1.2 - 2.11)
	Coeff b	-2.15	-0.71	- 4.8
Bleeding	Valor p	0.03	0.99	0.99
Ũ	OR (IC _{95%})	0.12 (0.11 – 1.2)	1.8 (0.3 – 2.9)	12.1 (7 - 14)
	Coeff b	-4.61	- 0.45	-1.77
Mortality	Valor p	0.004	0.4	0.99
	OR (IC _{95%})	0.8 (0.4 – 0.7)	1.57 (1.01 - 5)	5.85 (0.4 – 11.1)

Table 8. Logistic regression for the prognostic nutritional index (PNI), hemoglobin and prealbumin.

Due to the higher incidence of GC, Eastern countries have established screening programs, which leads to increased detection of neoplasms in early stages, with a higher volume of radical gastrectomies and lower surgical morbidity and mortality ²⁴. On the other hand, this study is limited because GC that is potentially curable with surgical management is not a very frequent entity in our setting.

In the Asian literature, mainly in Japan, several studies have been published ^{24–26} that, like ours, determine the PNI prior to gastrectomy and study the possible relationship between a decreased PNI and postoperative morbidity and mortality. Although the results obtained have not been conclusive, similarly to the results of our study, it is suggested that the presence of a low PNI could have a predictive value on the frequency of complications and overall survival. However, there is a limitation because it is known that there are other factors that also influence patient outcomes, such as chemotherapy and the psychological part, who-se analysis was not the objective of this study ²⁴.

By identifying predictive factors of surgical morbidity, individualized perioperative management could be provided, improving short- and long-term results in terms of disease-free period and overall survival of cancer patients ²⁴. In clinical practice, PNI can therefore be used to identify patients at risk of poor prognosis, predict sites of disease recurrence and monitor hematogenous metastases, and even define the indication for adjuvant chemotherapy in certain patients' stages in which there is still discussion.

Our results show that PNI is associated with mortality and postoperative complications in patients undergoing gastrectomy for gastric cancer, because PNI may reflect nutritional status and inflammatory and immune responses. A high preoperative PNI in patients with GC suggests that active perioperative nutritional support may be a new method to improve patient outcomes.

In a recent study ²⁴, the postoperative survival rate of patients undergoing radical gastrectomy with a PNI less than 46.03 was significantly lower than those with a PNI greater than or equal to 46.03 (23.7 months versus 34.3 months; p<0.001), results that contrast with ours. Despite the improvement in perioperative care and surgical techniques, R0 resection of the GC presents a high frequency of complications and maintains a significant mortality rate. Morbidity after radical gastrectomy is highly variable, with ranges between 10.5 and 46% and mortality varies from 0.6 to 10.2% ^{12,24,34}.

The vast majority of studies that analyze possible prognostic factors in GC refer to cases in which an apparently curative gastrectomy (RO) has been performed, so its usefulness is limited in the large percentage of patients in whom radical tumor resection is not performed. Currently, we lack sufficient data in our environment about the variables with prognostic value that can be used at the time of tumor diagnosis, which is when a therapeutic decision must be made ¹².

The limitations of this study arise from being a retrospective analysis, with a small sample size. Large-scale, multicenter, prospective studies should be designed. We only collected data on patient survival, but we did not have access to complete and effective data on disease-free survival, therefore it was not possible to analyze these important prognostic indicators.

Conclusions

The high frequency of deaths from GC throughout the world has led researchers to find strategies to reduce mortality. Few prognostic factors have been described in the literature that affect the survival of patients with GC, the main one being surgery, however, it continues to have a significant mortality rate and associated complications that reduce survival and patients' quality of life.

It is necessary to find anthropometric, clinical and paraclinical prognostic factors that allow establishing the risk in patients with GC to define the best surgical time and type of surgery; among these, nutritional status is a modifiable prognostic factor with treatment, which can influence both postoperative morbidity and survival.

The prognostic nutritional index (PNI) reflects the immunocompetent and nutritional status of

patients. Several studies, such as ours, have shown that patients with low preoperative PNI have poor survival, higher mortality, and postoperative complications when undergoing gastrectomy for gastric cancer.

The identification of predictive factors for surgical and post-surgical morbidity and the presentation of complications is the new challenge of gastric cancer research, in order to better guide individualized perioperative plans for each patient.

Compliance with ethical standards

Informed consent: The approval of the institutional ethics committee was obtained. The ethical aspects were adjusted to the fundamental ethical principles, to the guidelines of the Declaration of Helsinki, World Medical Association, Belmont Report, Guidelines issued by the World Medical Association and article 11 of Resolution 008430 of October 4, 1993 of the Ministry of Health of Colombia.

Telephone informed consent was requested from the patient or his relative when a telephone call was made to complete missing data in the patient's clinical history.

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

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Author's contributions:

- Conception and design of the study Andrés Eduardo Marín-Castro, Douglas Omar Ortiz-Espinel, Carlos Alberto Sánchez-Toro, Carlos Manuel Zapata-Acevedo, María José Marín-Castro, Blas Darío Conde-Rodríguez.
- Data acquisition: Andrés Eduardo Marín-Castro, María José Marín-Castro, Blas Darío Conde-Rodríguez.
- Data analysis and interpretation: Andrés Eduardo Marín-Castro, Douglas Omar Ortiz-Espinel, Carlos Alberto Sánchez-Toro, Carlos Manuel Zapata-Acevedo, María José Marín-Castro, Blas Darío Conde-Rodríguez, Gerardo Ardila-Duarte.
- Drafting the manuscript: Andrés Eduardo Marín-Castro, Douglas Omar Ortiz-Espinel, Carlos Alberto Sánchez-

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- Critical review: Douglas Omar Ortiz-Espinel, Carlos Alberto Sánchez-Toro, Carlos Manuel Zapata-Acevedo, Gerardo Ardila-Duarte.

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