Osmangazi Tıp Dergisi
Osmangazi Journal of Medicine 2023;

Research Article / Araştırma Makalesi

Factors Affecting Perioperative Period Renal Function in Nephrectomies

Nefrektomide Perioperatif Böbrek Fonksiyonunu Etkileyen Faktörler

[1]Meryem Onay, [1]Sema Sanal Baş, [2]Gizem Kurada, [3]Ata Özen, [1]Ebru Karakoç, [1]Birgül Yelken

[1]Eskisehir Osmangazi University, Faculty of Medicine, Department of Anesthesiology and Reanimation, Eskisehir, Turkey

[2]Kastamonu Training and Research Hospital, Department of Anesthesiology and Reanimation, Kastamonu, Turkey

[3]Eskisehir Osmangazi University, Faculty of Medicine, Department of Urology, Eskisehir, Turkey

Abstract

Patients who have had a nephrectomy usually have a history of renal dysfunction or are at risk for kidney failure due to tissue loss during surgery. In this study, our aim was to evaluate the factors affecting acute renal function in nephrectomy surgeries under general anesthesia. Demographic data of patients, case type, surgery type, duration of anesthesia, intraoperative lactate level, and postoperative complications of patients who underwent nephrectomy in our clinic were reviewed. Renal function was compared with urea, creatinine and estimated glomerular filtration rate (eGFR) levels in the preoperative (2 days before surgery) and postoperative period (day 2). The mean age of the patient was 58.2 ± 13.02 years, duration of anesthesia 166.24 ± 53.01/min, pre-operative eGFR 84.71 (67.69-90.0) mL/min/1.73 m2, and postoperative eGFR 65.09 (47.91-87.44) mL/min/1.73 m2 were respectively. When the pre- and postoperative renal functions of the patients were compared, the statistically significant results were those of urea (p: 0.01), creatinine (p:0.01), and GFR (p:0.01). The factors affecting GFR decrease were age (p:0.01) and case type (p:0.01). Increase in lactate levels in the postoperative period compared to the preoperative levels was statistically significant (p: 0.01). The increase in lactate levels was associated with complications (p: 0.001), case type (p:0.01) and surgery type (p: 0.01). Conclusions: The incidence of acute renal failure is increasing in nephrectomy surgeries, especially in radical ones. Age and case type are also the most important parameters to be consider.

Keywords: Nephrectomy, Renal function, Glomeruler filtration rate

Özet

Nefrektomi operasyonu geçiren hastalar genellikle öncesinde renal disfonksiyona sahip veya cerrahi sırasında meydana gelen doku kaybı nedeniyle renal yetmezlik açısından risk grubunda yer almaktadır. Bu çalışmada nefrektomi operasyonlarında genel anestezisi altında böbrek fonksiyonlarını etkileyen faktörleri değerlendirmeyi amaçladık. Kliniğimizde nefrektomi operasyonu geçiren hastaların demografik verileri, vaka türü (basit, parsiyel, radikal), laparotomi/laparoskopik, intraoperatif laktat düzeyi, anestezi süresi ve postoperatif komplikasyonlar kayıtları incelendi. Böbrek fonksiyonları preoperatif ve postoperatif dönemde (2. günde) üre, kreatinin ve tahmini glomeruler filtrasyon hızı (GFR) sonuçları ile karşılaştırıldı. Hastaların ortalama yaş 58.2 ± 13.02 iken, anestezi süresi 166.24 ± 53.01/dk, preoperatif eGFR 84.71 (67.69-90.0), postoperatif eGFR 65.09 (47.91-87.44) idi. Hastalar preoperatif ve postoperatif böbrek fonksiyon testleri kıyaslandığında; istatiksel olarak üre (p:0.01), kreatinin (p:0.01) ve GFR (p:0.01). GFR düşüşünü etkileyen faktörler, yaş (p:0.01) ve vaka türü. (p:0.01). Preoperatif döneme göre postoperatif dönemde laktat düzeylerindeki artış istatistiksel olarak anlamlıydı (p: 0.01). Laktat düzeyindeki artış komplikasyonlar (p: 0.001), vaka türü (p:0.01) ve cerrahi türü (p: 0.01) ile ilişkiliydi. Nefrektomi operasyonlarında özellikle radikal olanlarda kronik böbrek yetmezliği görülme sıklığı artmaktadır. Yaş ve vaka tipi de dikkate alınması gereken önemli parametrelerdir.

Anahtar Kelimeler: Nefrektomi, Böbrek fonksiyonu, Glomerular filtrasyon hızı

Correspondence:
Meryem ONAY
Eskisehir Osmangazi University, Faculty of Medicine, Department of Anesthesiology and Reanimation, Eskisehir, Turkey
e-mail: dr.meryemonay@hotmail.com

Received 16.01.2023 Accepted 20.02.2023 Online published 27.02.2023

Onay M, Sanal Bas S, Kurada G, Ozen A, Karakoc E, Yelken B. Factors Affecting Perioperative Period Renal Function in Nephrectomies, Osmangazi Journal of Medicine, 2023;45(3): 366-372 Doi: 10.20515/otd.1236019

1. Introduction

Nowadays, simple, partial, and radical nephrectomy surgeries are performed due to cancer, kidney injuries, and other diseases (1). Nephrectomy surgery results in parenchymal tissue loss, which contributes to acute or long-term renal function loss (2). Patients who have undergone nephrectomy constitute a risk group in terms of deterioration in renal function; hence, anesthesia technique and drugs used during the surgery are important (1,3). Generally, general anesthesia is preferred in nephrectomy considering surgical position and duration. In general anesthesia, inhalation anesthetics are used according to with the preference and experience of the anesthesiologist. Different inhalation anesthetics have effects on circulation, hemodynamics, and renal perfusion (4,5).

Decreased renal function has been emphasized as a cause of mortality in many studies. Glomerular filtration rate (GFR) is proportional to nephron reserve and is used to evaluate renal function in healthy individuals (6). In the literature, a GFR of <60 mL/min/1.73 m^2 has been associated with increased mortality, especially cardiovascular pathology. In society, the incidence of chronic kidney disease (CKD) has been increased due to diabetes, secondary to obesity, hypertension, and perivascular diseases. Existing comorbidities also negatively affect eGFR results in nephrectomy surgeries performed in long-term localized kidney tumors (7).

The aim of this study was to retrospectively evaluate the factors affecting renal function in a risk group with respect to renal dysfunction such as patients who underwent nephrectomy under general anesthesia.

2. Materials and Methods

After obtaining the approval of the ethics committee, numbered 2020-30, patients who underwent nephrectomy (simple, radical, partial) between January 2017 and February 2020 were included in the study. Patients who used volatile anesthetics other than sevoflurane were excluded from the study. Nephrectomy surgeries in our clinic are performed with laparotomy or laparoscopy. Age, sex, physical status classification of American Society of Anesthesiologists (ASA), history of pre-operative renal disease, case type (simple, partial, radical), surgery type (laparotomy/laparoscopy), duration of anesthesia, intraoperative lactate level and postoperative complications were examined. Renal functions of the patients were compared in the preoperative (2 day before surgery) and postoperative period (day 2) in terms of urea, creatinine, and eGFR results. With regard to anesthesia management, general anesthesia was applied to all patients by connecting them to a mechanical ventilation device after tracheal intubation. The anesthesia protocol was as follows: for induction, intravenous anesthetic (thiopental, propofol), muscle relaxant (rocuronium), and analgesic (remifentanil, fentanyl), and for maintenance, volatile anesthetic (sevoflurane) and remifentanil infusion (0.1–0.3 mcg/kg), air/oxygen combination were administered. The anesthesia was maintained at 2–4 L/min, although flow was not standard. Patients' fluid resuscitation and hemodynamic monitoring were routinely performed. Paracetamol (1g) and tramadol (1mg/kg) were administered as postoperative analgesia. In our clinic, Dräger Perseus® A500 closed circuit system anesthesia devices, where the carbon dioxide absorbent is soda lime, are used.

Statistical analysis

All data were recorded in the SPSS (statistical package for social sciences) for Windows 22 program on the computer and analyzed. In the analysis of the data, firstly, the assumptions that must be met in order to decide which tests (parametric / nonparametric tests) to apply are tested. In order to decide the normality of the distribution, Kolmogorov-Smirnov test, kurtosis and skewness values, which are other assumptions of normal distribution, and histogram plot were used. Summary values of quantitative (numerical) variables are shown as mean±S.D, median and inter-quartile difference, while summary values for qualitative (categorical) variables are shown as frequency and percentage. Man Whitney-U test and Kruskal Wallis-H test were used for

comparing two independent groups, and Wilcoxon test was used to compare two related groups. The relationship between the variables was analyzed using the Spearman correlation coefficient. The significance level of 0.05 was used as the criterion in interpreting the significance of the obtained values.

3. Results

In this study, we evaluated 185 patients who underwent nephrectomy. The mean age of the patients and duration of anesthesia were 58.2 ± 13.02 years and 166.24 ± 53.01 min, respectively. Of the patients sex and the American Society of Anesthesiologists physical status classification system in table 1. In the pre-operative evaluation, 11 of the patients had renal disease (3 patients were dependent on hemodialysis treatment) (Table 1).

The type of nephrectomy and a surgical method performed in table 1. When the pre- and postoperative renal function of the patients were compared, the statistical significance of the results was as follows in table 2.

The relationship between the decrease in the post- and pre-operative urine levels and sex, age, case type, and ASA; the relationship between the increase in creatinine and age, case type; the relationship between the decrease in eGFR and age, case type were evaluated as statistically significant. Details in the table 3-4.

Increase in lactate levels in the postoperative period 1.3 mmol/L (1-1.8) compared to the pre-operative levels 0.9 mmol/L (0.7-1.3) was statistically significant (p: 0.01). The increase in lactate levels was not statistically significant in relation to age, sex and ASA; however, it was associated with complications (p: 0.001), case type (p:0.01) and surgery type (p: 0.01).

The peri-operative complications are intraoperative bleeding (n:13), angina (n:1), nausea and vomiting (n:1), respiratory distress (n:5), signs of infection (lung infection, fever, urinary tract infection, wound infection) (n:8), postoperative hemodialysis (n:2). Cardiac arrest secondary to intraoperative hemorrhage developed in two patients in the postoperative days. The most common complication was intraoperative hemorrhage. It was observed in radical nephrectomies (n:8), partial nephrectomy (n:4) and simple nephrectomy (n:1) according to the extent of surgery.

Table 1. Demographic data and case information

Parameters	**All (n: 185)**
Age (year)	58.2 ± 13.02
Sex	
Male	107 (57.8%)
Female	78 (42.2%)
American Society of Anesthesiologists (ASA) physical status classification	
ASA I	
ASA II	n: 40 (21.6%)
ASA III	n: 109 (58.9%)
	n: 36 (19.5%)
Pre-operative renal disease	n: 11 (5.9%)
(n:3 hemodialysis treatment dependent)	
Case type	
Simple	n: 33 (17.8%)
Partial	n: 80 (43.2%)
Radical	n: 72 (38.9%)
Surgery type	
Laparotomy	n:159 (85.9%)
Laparoscopy	n: 26 (14.1%)
Duration of anesthesia (min)	166.24 ± 53.01

Data are expressed as n (%) or mean ± STD. ASA: American Society of Anesthesiologists.

Table 2. Evaluation of pre-operative and postoperative renal function

	Pre-operative median (Q1-Q3)	Postoperative median (Q1-Q3)	p
Urea (mg/dL)	15.0 (12.00-19.00)	13.00 (10.00-18.00)	0.01*
Creatinine (mg/dL)	0.89 (0.74-1.09)	1.07 (0.88-1.48)	0.01*
GFR (ml/dk)	84.71 (67.69-90.00)	65.09 (47.91-87.44)	0.01*
Lactate (mmol/L)	0.9 (0.7-1.3)	1.3 (1-1.8)	0.01*

Table 3. Relationship of age with postoperative and preoperative change in GFR

	Age	
	r	**p**
Urea (mg/dL)	0.24	0.01*
Creatinine (mg/dL)	0.30	0.01*
GFR (ml/dk)	-0.30	0.01*
Lactate (mmol/L)	-.004	0.96

** $p < 0.05$ was considered statistically significant.*
GFR: Glomerular filtration rate

Table 4. Effect of case type on postoperative and preoperative change in GFR

	Radical Median (Q1-Q3)	Partial Median (Q1-Q3)	Simple Median (Q1-Q3)	p
Urea mg/dL)	(-0.35) (-2.60-3.23)	(-1.45) (-4.00-2.08)	(-4.2) (-7.7 -2.85)	0.01*
Creatinine mg/dL)	0.42 (0.17-0.57)	0.14 (0.03- 0.37)	0.1 (-0.1-0.06)	0.01*
GFR (ml/dk)	(-23.29) (-30.99-8.26)	(-8.76) (-24.22- 0.0)	(0.01) (-2.2-4.6)	0.01*
Lactate (mmol/L)	0.20(0.0-0.4)	0.4(0.2-0.6)	0.05(0-0.3)	0.01*

** $p < 0.05$ was considered statistically significant.*
GFR: Glomerular filtration rate

4. Discussion

In patients who have undergone kidney surgery, usually with renal dysfunction prior to surgery or tissue loss during surgery, loss of renal function with acute and chronic processes may develop. Sevoflurane is frequently preferred due to its fragrant, less irritant, and bronchodilator effect advantages compared to other agents (4). Volatile agents used in anesthesia are reduced to toxic byproducts by carbon dioxide absorbents (8). It has been reported, especially in dose-dependent animal studies, that sevoflurane induces nephrotoxicity through compound A (9,10). Although there is no clinical evidence of nephrotoxicity with volatile anesthetics currently used in general anesthesia, it is important for the protection of renal function in the postoperative period in patients undergoing nephrectomy. In this study, we retrospectively examined the cases of nephrectomy in our clinic in the last 3 years. A total of 185 patients who received sevoflurane as a volatile anesthetic were included in the study. When renal function tests were evaluated pre-operatively (2 day before surgery) and on the second postoperative day, a significant change was observed in urine levels, creatinine and eGFR levels.

Acute kidney injury (AKI) is a common complication in cardiac surgeries. Although animal studies have shown that volatile anesthetics have a protective effect, the clinical effect is not clear. It has been associated with volatile anesthetics providing

better hemodynamic stability compared to other techniques and with a decrease in the requirement of inotropic agents (11). In some studies, eGFR has been preferred as a biomarker for evaluating renal functions. Although serum creatinine level is a simpler and cheaper parameter in practical use, it is affected by individual conditions (age, muscle mass, medication, etc.) (12). At the same time, differences were found in AKI, which develops due to ischemia-reperfusion injury in many organs including kidney. It was observed that the female sex, especially those under the age of 52, was protected in AKI (13). In our study, the increase in creatinine observed perioperatively was smaller in women, but it was not statistically significant with gender (p:0.079). There was a significant decrease in eGFR, but it was not observed to be related to sex. Ultimately, although less specific than renal biomarkers, eGFR was relevant for clinical research as it is more practical.

Partial nephrectomy is the gold standard method in small kidney tumors. Partial nephrectomy causes less renal dysfunction than radical nephrectomy, but is at risk due to parenchymal loss and damage during surgery. During abdominal surgery, renal blood flow also decreases due to surgery and anesthesia. To reduce parenchymal bleeding and improve visibility in the surgical field, clamping is performed in the renal vessels. The resulting ischemia reperfusion is considered a risk factor for acute renal damage (2,14). In our study, partial nephrectomy was performed in 43.2 % of the patients. In partial nephrectomies, there was a significant decrease in the pre-operative 83.49 ± 15.10 and postoperative eGFR 70.82 ± 22.83 (p:0.01).

Radical nephrectomy surgery is commonly used in renal cell carcinoma. Due to pre-existing renal parenchymal changes, a decrease in renal function is inevitable and poses a risk for newly-developing CKD (15,16). A new-onset CKD has adverse outcomes, including an increased risk of cardiovascular disease and death. Brandina et al. (15) emphasized the effectiveness of the Charlson comorbidity index, baseline GFR, and global glomerulosclerosis rate. It was observed that there was a 26.5% decrease in GFR during a mean follow-up of 49 months in patients who underwent radical nephrectomy. For new-onset CKD, eGFR was assumed as <60. Among 35 patients in the long-term follow-up, 53.8% developed new-onset kidney disease (15). In our study, the pre-operative eGFR was 73.8 ± 21.24 in radical nephrectomies, while the postoperative eGFR was 53.4 ± 20.84, corresponding to a 27.6% decline. Our data included a 48-hour postoperative acute period, and our rates were similar compared to the study.

Radical nephrectomy is technically easier than partial nephrectomy and associated with fewer complications. Compared to partial nephrectomy, it also eliminates oncological concerns in terms of resection margins. However, in radical nephrectomies, there is a higher risk of developing renal failure than partial nephrectomies. It has been shown that age, pre-operative creatinine, and pre-operative eGFR were affecting factors (17). In our study, in patients with partial nephrectomy, the pre-and postoperative eGFR were 83.49 ± 15.10 and 70.82 ± 22.83, respectively, % 15.2 decrease was observed. The risk of acute renal failure is increased in radical nephrectomies compared to partial ones.

Recently, renal disease has often been reported to be associated with hypertension and diabetes mellitus in the society (18). In other words, it should be kept in mind that surgery is not the only factor in patients undergoing nephrectomy and may affect renal functions in comorbidities. In our study, it was observed that in the ASA scoring independent of surgery where comorbidities of the patients were evaluated, the changes in levels of urea was statistically significant. It was observed that the factor of increased age affected urea, creatinine, and GFR levels.

Duymaz et al. compared renal functions in patients who were given low-flow sevoflurane and desflurane anesthesia in urological surgeries, and it was observed that there was no difference between the groups. In evaluation of renal functions, cystatin C,

which is more sensitive than creatinine, creatinine, and creatinine clearance were used to show early and mild changes. As a result, it was shown that sevoflurane is safe in terms of renal function in low-flow anesthesia (19). Urological surgeries involve pathological diseases of the urinary system; however, in our study, we preferred to evaluate nephrectomies, a more specific factor that results in tissue loss, in terms of renal function. Although low-flow anesthesia was not used, the change in creatinine and eGFR was significant in the pre- and postoperative periods, and we wanted to emphasize that the only factor would not be the use of volatile anesthetic.

In the study conducted by Ebert et al. wherein genitourinary system surgeries and patients with renal insufficiency were excluded, it was revealed that the changes in renal functions did not depend on the anesthetic agent used. It has been suggested that the antibiotic agent used, duration of surgery, width of the site of surgery, diffuse stress, existing renal dysfunction, and pre-operative blood pressure changes may affect renal function (20). Considering the results of our study, when the volatile agent was kept constant, it was seen that case type and age affected eGFR and creatinine values. Intraoperative bleeding accompanied by haemodynamic changes constitute the majority of perioperative complications. The significant increase in postoperative lactate levels compared to preoperative levels supports its relation with complications, case type and surgery type.

The limitations of the study were the retrospective nature of data collection, single-center nature of the study, and uneven distribution among the patient groups. Renal function was not analyzed by specific tests; it was evaluated by routine laboratory tests. Although pre- and postoperative renal function parameters and demographic data were evaluated, short-term records were reviewed in terms of renal function. Parameters that may affect other renal functions including intraoperative bleeding, urine output amount, hypertension, heart failure, and comorbidities such as benign prostatic hyperplasia were not evaluated.

5. Conclusion

Patients undergoing nephrectomy have an increased risk of chronic kidney disease due to pre-existing renal problems and existing comorbidities. We evaluated renal function using eGFR in the acute postoperative period. We concluded that advanced age and case type are effective in decreasing eGFR, especially in radical nephrectomy.

REFERENCES

1. Mishra A, Verma R, Bhatia V, et al. Thoracic Paravertebral Block for Postoperative Pain Management in Patients Undergoing for Nephrectomy: A Randomised Clinical Trial. *Journal of Clinical & Diagnostic Research.* 2018;12(10).
2. Lane BR, Babitz SK, Vlasakova K, et al. Evaluation of Urinary Renal Biomarkers for Early Prediction of Acute Kidney Injury Following Partial Nephrectomy: A Feasibility Study. *European urology focus.* 2020;6:1240-1247.
3. Lee HT, Ota-Setlik A, Fu Y, Nasr SH, Emala CW. Differential protective effects of volatile anesthetics against renal ischemia–reperfusion injury in vivo. Anesthesiology: *The Journal of the American Society of Anesthesiologists.* 2004;101:1313-24.
4. Yildirim M, Kucuk H, Demir T, et al. Early Allograft Function in Renal Transplant Recipients: Is it Affected by Volatile Anesthetics? Transplantation Proceedings; 2015: Elsevier.
5. Ko JS, Gwak MS, Choi SJ, et al. The effects of desflurane and sevoflurane on hepatic and renal functions after right hepatectomy in living donors. *Transplant International.* 2010;23:736-44.
6. Gazel E, Biçer S, Ölçücüoğlu E, et al. Comparison of renal function after donor and radical nephrectomy. *Renal Failure.* 2015;37:377-80.
7. Pettus JA, Jang TL, Thompson RH, Yossepowitch O, Kagiwada M, Russo P, editors. Effect of baseline glomerular filtration rate on survival in patients undergoing partial or radical nephrectomy for renal cortical tumors. Mayo Clinic Proceedings; 2008: Elsevier.
8. Marini F, Bellugi I, Gambi D, et al. Compound A, formaldehyde and methanol concentrations during low-flow sevoflurane anaesthesia: comparison of three carbon dioxide absorbers. *Acta anaesthesiologica scandinavica*. 2007;51:625-32.

9. Conzen PF, Kharasch ED, Czerner SF, et al. Low-flow sevoflurane compared with low-flow isoflurane anesthesia in patients with stable renal insufficiency. Anesthesiology: *The Journal of the American Society of Anesthesiologists*. 2002;97:578-84.

10. Fukuda H, Kawamoto M, Yuge O, Fujii K. A comparison of the effects of prolonged (> 10 hour) low-flow sevoflurane, high-flow sevoflurane, and low-flow isoflurane anaesthesia on hepatorenal function in orthopaedic patients. *Anaesthesia and intensive care.* 2004;32:210-8.

11. Cai J, Xu R, Yu X, Fang Y, Ding X. Volatile anesthetics in preventing acute kidney injury after cardiac surgery: a systematic review and meta-analysis. *The Journal of thoracic and cardiovascular surgery*. 2014;148:3127-36.

12. Sindhvananda W, Phisaiphun K, Prapongsena P. No renal protection from volatile-anesthetic preconditioning in open heart surgery. *Journal of anesthesia*. 2013;27:48-55.

13. Kher A, Meldrum KK, Wang M, et al. Cellular and molecular mechanisms of sex differences in renal ischemia–reperfusion injury. *Cardiovascular research.* 2005;67:594-603.

14. Toprak H, Şahin T, Aslan S, et al. editors. Effects of desflurane and isoflurane on hepatic and renal functions and coagulation profile during donor hepatectomy. Transplantation proceedings; 2012: Elsevier.

15. Brandina R, Leite KRM, Gregório EP, Fernandes KBP, Srougi M. Histologic abnormalities in non-neoplastic renal parenchyma and the risk of chronic kidney disease following radical nephrectomy. *Urology.* 2017;100:158-62.

16. Yokoyama M, Fujii Y, Iimura Y, et al. Longitudinal change in renal function after radical nephrectomy in Japanese patients with renal cortical tumors. The Journal of urology. 2011;185:2066-71.

17. Choi YS, Park YH, Kim Y-J, et al. Predictive factors for the development of chronic renal insufficiency after renal surgery: a multicenter study. *International urology and nephrology.* 2014;46:681-6.

18. Bakris GL, Williams M, Dworkin L, et al. Preserving renal function in adults with hypertension and diabetes: a consensus approach. *American journal of kidney diseases.* 2000;36:646-61.

19. Duymaz G, Yağar S, Özgök A. Comparison of effects of low-flow sevoflurane and low-flow desflurane anaesthesia on renal functions using cystatin C. *Turkish Journal of Anaesthesiology and Reanimation.* 2017;45:93.

20. Ebert TJ, Arain SR. Renal responses to low-flow desflurane, sevoflurane, and propofol in patients. Anesthesiology: *The Journal of the American Society of Anesthesiologists.* 2000;93:1401-6.

Ethics

Ethics Committee Approval: The study was approved by Eskişehir Osmangazi University Noninterventional Clinical Research Ethical Committee (Decision no:30, Date: 12.05.2020).

Informed Consent: The authors declared that it was not considered necessary to get consent from the patients because the study was a retrospective data analysis.

Authorship Contributions: Conceptualization, MO, SBS Data curation, GK,SSB,EK; Investigation, EK; Methodology, MO, AÖ, BY; Supervision, BY, SSB, EK; Writing–original draft, MO; Writing–review & editing, MO, BY.

Copyright Transfer Form: Copyright Transfer Form was signed by all authors.

Peer-review: Internally peer-reviewed.

Conflict of Interest: No conflict of interest was declared by the authors.

Financial Disclosure: The authors declared that this study received no financial support.