

RESEARCH ARTICLE

Effect of Preoperative and Postoperative Erector Spinae Plane Block on Perioperative Hemodynamics and Postoperative Analgesia in Video-assisted Thoracoscopic Surgery: A Randomized Controlled Study

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ABSTRACT

Objectives: To compare the hemodynamic and analgesic effects of pre-/postoperative erector spinae plane block (ESPB) application in patients undergoing video-assisted thoracoscopic surgery (VATS).

Methods: This was a prospective, randomized multicenter study. Patients were assigned to preoperative ESPB (Group-Pre; n=32) or postoperative ESPB (Group-Post; n=33) groups. Ultrasound-guided block applications were performed under general anesthesia with single-needle insertion. Pain scores were assessed by visual analog scale (VAS). Demographic characteristics and surgical procedure data of the patients were recorded. In addition, perioperative mean arterial pressure (MAP), peripheral oxygen saturation (SpO₂), heart rate (HR), and bispectral index (BIS) values were recorded. MAP, SpO₂, HR, VAS scores (while resting/coughing), additional analgesic use, morphine consumption, and side effects were recorded 24 hours postoperatively.

Results: The groups were statistically similar in terms of MAP, HR, and SpO_2 in the intraoperative and postoperative periods. VAS resting and coughing values were statistically significantly higher at the 1st, 2nd, 4th, and 12th hours in Group-Post compared with Group-Pre (p<0.05). There was no statistically significant difference between the groups in terms of 24-hour VAS at rest (p=0.258) or VAS at cough (p=0.189). The amount of remifentanil requirement, morphine consumption, and additional analgesic use in Group-Post was statistically significantly higher than in Group-Pre (p<0.05).

Conclusion: ESPB applied in the preoperative period is more effective in suppressing the surgical response in VATS, as it limits intraoperative opioid consumption and provides more effective analgesia in the postoperative period.

Keywords: Acute pain, erector spinae plane block, hemodynamics, postoperative pain, video-assisted thoracoscopic surgery

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Introduction

Video-assisted thoracoscopic surgery (VATS) is a minimally invasive technique that provides faster recovery after thoracic surgery.^[1,2] However, conditions that may occur during

VATS, such as intercostal nerve, muscle, rib retraction, and pleural damage, can cause severe and long-lasting pain.^[3,4]

A multimodal analgesia approach for postoperative pain control after VATS includes the use of local anesthetics

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with thoracic epidural analgesia and chest wall blocks such as intercostal block, erector spinae plane block (ESPB), serratus anterior plane block, and paravertebral block.^[5] In VATS applications, chest wall blocks are a component of Enhanced Recovery After Surgery (ERAS) protocols and are also recommended by the European Society of Regional Anaesthesia.^[6,7]

ESPB is an interfascial plane block defined by Forero et al.^[8] in 2016. Areas affected by ESPB include the dorsal and ventral branches of the spinal nerve and the lateral cutaneous branches of the intercostal nerves in the paravertebral space through the penetration of intertransverse connecting tissues.^[4] ESPB can be applied preoperatively or postoperatively.^[9,10]

General anesthesia (GA) is required in many thoracic surgical procedures. However, although GA produces anesthesia with only a central effect, it cannot completely prevent the transmission of peripheral painful stimuli to the central nervous system. This may result in a more significant intraoperative stress response. In addition, systemic opioid use causes delayed recovery, respiratory depression, and nausea/vomiting.^[11]

Preemptive analgesia applications via regional anesthesia provides a dense afferent blockade, abolishing somatosensory evoked potentials, and they block transmission within the sympathetic chain. Although many studies have demonstrated this situation, this issue remains unclear.^[12,13] ESPB is an interfascial plane block that targets both the ventral and dorsal branches of the spinal nerves.^[14] Preoperative ESPB applications added to GA in VATS operations lead to lower pain levels, reduced opioid consumption, low additional analgesia requirement, and fewer side effects.^[4,9] In addition, we did not find any study investigating the effect of ESPB application time (preoperative/postoperative) on intraoperative hemodynamics and postoperative analgesia.

In this study, we hypothesized that more stable hemodynamics and more effective postoperative analgesia could be achieved by providing preemptive analgesia with preoperatively applied ESPB in patients undergoing VATS. For this purpose, we compared the effects of preoperative and postoperative ESPB applications in patients who underwent VATS.

Methods

Study Design and Patients

The study was conducted in two centers (Ankara Bilkent City Hospital and Ankara Atatürk Sanatorium Training and Research Hospital). It was planned as a prospective, randomized, single-blind design. The study was approved by the Ankara City Hospital Institutional Review Board (E. Kurul-E1-22-2536/04.2022) and registered on clinicaltrials. gov (NCT05334628).

Between April 2022 and September 2022, patients aged 18–80 years, with a body mass index (BMI) of 18–40 kg/m², American Society of Anesthesiologists (ASA) classification of 1–3, and undergoing VATS were included in the study. All patients were informed about the study, and their written/ verbal consent was obtained. Patients who were included in the study were also given pain assessment and patient-controlled analgesia (PCA) training.

Patients with a history of a bleeding disorder, chronic pain treatment, local anesthetic allergy, infection in the area to be blocked, converted to thoracotomy, and operated under emergency conditions were not included in the study. The primary outcome was determined as perioperative mean arterial pressure. Secondary outcomes were perioperative heart rates, peripheral oxygen saturation (SpO₂), postoperative visual analog scale (VAS) scores, additional analgesic requirements, morphine consumption, and intraoperative bispectral index (BIS) values.

The patients were divided into the preoperative ESPB group (Group-Pre) and the postoperative ESPB group (Group-Post). Using computer-generated random numbers, the patients were randomly assigned to two groups of 30 individuals each, with an allocation ratio of 1:1.

General Anesthesia

After standardized monitoring by ASA, the patients were given standard 0.03 mg/kg intravenous (IV) midazolam before the operation. After preoxygenation, anesthesia was induced with 2 mg/kg propofol, 1.5 μ g/kg fentanyl, and 0.1 mg/kg vecuronium. After the patients were administered neuromuscular blockade, they were intubated with an appropriately sized left double lumen tube. Tube control was achieved with fiber-optic bronchoscopy. Anesthesia was maintained with sevoflurane and remifentanil infusion (0.01–0.20 μ g/kg/min). The remifentanil infusion dose was determined according to hemodynamic changes. Biportal VATS was applied to all patients, and a single chest tube was inserted.

Block Procedures

After strict sterile conditions were provided, we performed ultrasound (US)–guided block applications under GA in the lateral decubitus position. A linear probe and a US-compatible 22-gauge and 8-mm nerve block needles were used for the block application. The US probe was placed in plane 2 cm lateral to the T5 transverse process. Location accuracy was achieved via hydrodissection with 2 mL of saline. We then injected 30 mL of 0.25% bupivacaine into this area.



Figure 1. Flowchart of the patients.

Group-Pre (n=30): ESPB was applied to these patients under GA in the lateral decubitus position before the surgical incision.

Group-Post (n=30): ESPB was applied to these patients under GA in the lateral decubitus position at the end of the surgery, just before awakening.

Analgesia Protocol

We administered IV 100 mg of tramadol and 50 mg of dexketoprofen to the patients for multimodal analgesia at the end of the surgery. We administered IV 10 mg of metoclopramide to prevent nausea/vomiting. In the postoperative period, IV PCA prepared with morphine was applied. The PCA pump dose delivery was limited to administering a bolus dose of 1 mg of morphine and delivering a maximum dose of 12 mg of morphine in total within 4 h with lockout intervals of 15 minutes. In addition, 50 mg dexketoprofen every 12 hours and 1 g acetaminophen every 8 hours were ordered. Pain was defined with a 0- to 10-point (0: no pain and 10: unbearable pain) VAS. When the VAS score was ≥4, we administered 0.5 mg/kg of tramadol via IV for rescue analgesia. Patients who were followed up in the surgical intensive care unit for 24 hours were then transferred to the surgical service. Block applications were performed by two anesthesiologists experienced in the use of US in both centers. VAS follow-ups of patients were performed by pain management nurses who were blinded to the type of block applied to the patient. We recorded age, gender, BMI, and surgical procedure data of

the patients. In addition, perioperative (preanesthesia, presurgical incision, 5th, 30th, 60th, 90th, and 120th minutes after surgical incision) MAP, SpO₂, HR, and BIS values were recorded. In addition, MAP, SpO₂, HR, VAS scores at rest and while coughing, additional analgesia needs, morphine consumption, and side effects (hypotension, respiratory depression, nausea/vomiting, allergic reaction/itching, and urinary retention) were recorded at the postoperative 1st, 2nd, 4th, 12th, and 24th hours.

Statistical Analysis and Sample Size

Data analyses were performed using SPSS for Windows version 22.0 (SPSS Inc., Chicago, IL, United States). We used the Kolmogorov–Smirnov test to determine whether the distribution of continuous variables was normal. The Levene test was used to evaluate the homogeneity of variances. Unless specified otherwise, continuous data were described as mean±SD for normal distributions and median (Q1: first quartile – Q3: third quartile) for skewed distribu-

Table 1. Demographic and clinica	I characteristics of the patients

	Group-pre (n=30)		Group-post (n=30)		р
	n	%	n	%	
Age, year	49.57	7±18.20	49.47	7±16.97	0.993*
Gender					
Female	8	26.7	7	23.3	0.766 ^δ
Male	22	73.3	23	76.7	
BMI (kg/m²)	24.0	0±6.38	26.3	8±4.75	0.480*
Duration of anesthesia (minute)	180 (1	50–210)	173 (1	30–210)	0.468 ^β
Surgery					
Wedge	18	60.0	19	63.3	0.999 ^δ
Segmentectomy	1	3.3	-	-	
Lobectomy	11	36.7	11	36.7	
ASA					
ASAT	-	_	1	3.3	0.999 ^δ
ASATI	19	63.3	19	63.3	
ASATT	11	36.7	10	33.3	

Continuous variables are expressed as either * the mean \pm standard deviation (SD) or β the median (Q1: first quartile – Q3: third quartile), and categorical variables are expressed as either δ frequency or percentage. Continuous variables were compared with a Student t-test or the Mann-Whitney U test, and categorical variables were compared using Pearson's chi-square test or Fisher's exact test. BMI: Body mass index; ASA: American Society of Anesthesiologists.

MAP (mmHg)	Group-pre (n=30)	Group-post (n=30)	p *	SpO ₂ (%)	Group-pre (n=30)	Group-post (n=30)	Ρ ^β
Pre-anesthesia	99.17±11.55	98.43±16.09	0.840	Pre-anesthesia	97 (96–98)	97 (96–98)	0.052
Pre-incision	83.23±10.99	80.70±12.61	0.410	Pre-incision	99 (98–99)	98.5 (98–99)	0.095
5 th minutes	88.37±11.20	92.43±15.48	0.249	5 th minutes	99 (98–100)	98 (97–99)	0.164
30 th minutes	82.53±11.78	80.23±11.80	0.453	30 th minutes	99 (98–99)	98 (96–100)	0.459
60 th minutes	82.17±8.39	80.03±12.36	0.437	60 th minutes	99 (98–99)	98 (97–99)	0.421
90 th minutes	80.00±7.91	77.50±12.15	0.349	90 th minutes	99 (98–100)	98 (97–99)	0.090
120 th minutes	81.13±8.84	80.20±10.47	0.711	120 th minutes	99 (98–100)	99 (97–99)	0.092
Post-anesthesia	89.40±13.00	89.67±13.56	0.938	Post-anesthesia	98 (97–99)	98 (97–99)	0.874
HR (beat/min)	Group-pre	Group-post	р*	BIS values	Group-pre	Group-post	Ρβ
	(n=30)	(n=30)			(n=30)	(n=30)	
Pre-anesthesia	(n=30) 85.70±11.90	(n=30) 81.60±11.33	0.177	Pre-anesthesia	(n=30) 94 (92–95)	(n=30) 94 (92–97)	0.541
Pre-anesthesia Pre-incision			0.177 0.143	Pre-anesthesia Pre-incision			0.541 0.175
	85.70±11.90	81.60±11.33			94 (92–95)	94 (92–97)	
Pre-incision	85.70±11.90 80.80±10.67	81.60±11.33 76.57±11.40	0.143	Pre-incision	94 (92–95) 42 (41–43)	94 (92–97) 42 (40–42)	0.175
Pre-incision 5 th minutes	85.70±11.90 80.80±10.67 80.10±10.64	81.60±11.33 76.57±11.40 79.87±13.35	0.143 0.941	Pre-incision 5 th minutes	94 (92–95) 42 (41–43) 42 (41–43)	94 (92–97) 42 (40–42) 45 (42–48)	0.175 0.003
Pre-incision 5 th minutes 30 th minutes	85.70±11.90 80.80±10.67 80.10±10.64 78.67±11.80	81.60±11.33 76.57±11.40 79.87±13.35 75.83±12.55	0.143 0.941 0.371	Pre-incision 5 th minutes 30 th minutes	94 (92–95) 42 (41–43) 42 (41–43) 42 (41–44)	94 (92–97) 42 (40–42) 45 (42–48) 42 (41–43)	0.175 0.003 0.455
Pre-incision 5 th minutes 30 th minutes 60 th minutes	85.70±11.90 80.80±10.67 80.10±10.64 78.67±11.80 77.83±12.47	81.60±11.33 76.57±11.40 79.87±13.35 75.83±12.55 74.87±14.19	0.143 0.941 0.371 0.393	Pre-incision 5 th minutes 30 th minutes 60 th minutes	94 (92–95) 42 (41–43) 42 (41–43) 42 (41–44) 42 (41–44)	94 (92–97) 42 (40–42) 45 (42–48) 42 (41–43) 42 (40–43)	0.175 0.003 0.455 0.522

Table 2. Comparison of the groups in terms of MAP, HR, SpO₂, and BIS values in the intraoperative period

Continuous variables are expressed as either * the mean±standard deviation (SD) or β the median (Q1: first quartile – Q3: third quartile). Continuous variables were compared with a Student t-test and with the Mann-Whitney U test. Statistically significant p-values are in bold. BIS values in 5th minutes. MAP: Mean arterial pressure; HR: Hearth rate; SpO₂: Oxygen saturation; BIS: Bispectral index.

tions. Categorical data were described as many cases (%). We used the Student's t-test to compare statistical analysis differences in normally distributed variables between two independent groups and applied the Mann–Whitney U-test for comparisons of nonnormally distributed data. We compared categorical variables using Pearson's chisquare test or Fisher's exact test; a p value of <0.05 was accepted as significant in all statistical analyses.



Figure 2. Comparison of the groups in terms of MAP, HR, SpO₂, and BIS values in the intraoperative period. *p=0.003. MAP: Mean arterial pressure; HR: Heart rate; SpO₂: Oxygen saturation; BIS: Bispectral index.

We calculated the sample size using G*Power© software version 3.1.9.2 (Institute of Experimental Psychology, Heinrich Heine University, Dusseldorf, Germany). The sample size was calculated for the Mann–Whitney U-test, which was used to test the main hypothesis of (intraoperative MAP in the fifth minute) in the preliminary study. Based on the results of the preliminary study research using a two-sided (two-tailed) type I error of 0.05, power of 90% $(1 - \beta = 0.9)$, and effect size (d) factor of 0.93, we determined that ≥52 subjects were required for this study.

Results

Between April and September 2022, 65 patients from two centers who met the criteria participated in the study. We excluded five patients who converted to thoracotomy (Fig. 1). The patients participating in the study were similar in terms of demographic and clinical characteristics (Table 1). The groups were statistically similar in terms of MAP, HR, and SpO_2 at all intraoperative times. When compared in terms of BIS values, there was a statistically significant difference at the fifth minute in Group-Post. There was no statistically significant difference between the two groups in terms of BIS values at the other times (Table 2, Fig. 2).

There was no statistically significant difference between the groups in MAP, HR, or SpO₂ at all times in the postoperative period (Table 3).

The VAS resting values were statistically significantly higher at the 1st, 2nd, 4th, and 12th hours in Group-Post as compared with Group-Pre (p=0.027, p=0.003, p=0.001, and p<0.001, respectively). There was no statistically significant difference between the groups in 24th-hour VAS at rest (p=0.258). The 1st-hour, 2nd-hour, 4th-hour, and 12th-hour VAS coughing values were statistically significantly higher in Group-Post than in Group-Pre (p=0.024,

Tablo 3. Comparison of the groups in terms of MAP, HR, and ${\rm SpO}_{\rm 2}$ in the postoperative period

MAP (mmHg)	Group-pre (n=30)	Group-post (n=30)	р
1 st hour	89.53±10.50	94.23±11.20	0.099*
2 nd hour	87.47±10.74	91.13±9.59	0.169*
4 th hour	85.67±7.97	88.90±6.19	0.085*
12 th hour	84 (80-88)	88 (82–93)	0.154 ^β
24 th hour	89.03±14.36	88.33±11.61	0.836*
HR (beat/min)	Group-pre (n=30)	Group-post (n=30)	р
1 st hour	77.60±13.52	80.47±12.36	0.395*
2 nd hour	76 (70–80)	78 (74–88)	0.115 ^β
4 th hour	75 (72–82)	79 (72–84)	0.264 ^β
12 th hour	76 (72–82)	78 (74–81)	0.256 ^β
24 th hour	88.47±11.88	87.37±11.74	0.720*
SpO ₂ (%)	Group-pre (n=30)	Group-post (n=30)	р
1 st hour	98 (97–99)	98 (97–98)	0.203 ^β
2 nd hour	98 (97–99)	98 (97–98)	0.175 ^β
4 th hour	99 (98–99)	98 (96–98)	0.079 ^β
12 th hour	98 (97–99)	98 (97–98)	0.162 ^β
24 th hour	98 (96–98)	98 (96–98)	0.402 ^β

Continuous variables are expressed as either * the mean \pm standard deviation (SD) or β the median (Q1: first quartile – Q3: third quartile). Continuous variables were compared with a Student t-test and with the Mann-Whitney U test. MAP: Mean arterial pressure; HR: Hearth rate; SpO.; Oxygen saturation.

p=0.003, p=0.002, and p=0.002, respectively). There was no statistically significant difference between the groups in 24th-hour VAS at cough (p=0.189; Table 4).

The amount of remifentanil use, morphine consumption, and additional analgesia use in Group-Post were statistically significantly higher than in Group-Pre (p=0.001, p<0.001, and p<0.001, respectively). There was no statistically significant difference in nausea and vomiting between the groups (Table 5).

Discussion

The results of this study, in which we compared the preoperative and postoperative application of ESPB in VATS, showed that there was no difference in terms of vital parameters, but positive results were observed in the preoperative ESPB application, especially in terms of intraoperative opioid consumption and postoperative pain parameters.

In recent years, approaches to postoperative recovery (ERAS), including shortening the preoperative fasting period, multimodal analgesia combined with regional blocks, reduction of opiate use, early postoperative oral intake, early mobilization, and optimal pain control to prevent

	Group-pre (n=30)	Group-post (n=30)	р
VAS at rest			
1 st hour	2 (1–3)	4 (1–4)	0.027
2 nd hour	2 (0–2)	3 (1–4)	0.003
4 th hour	1 (0–2)	3 (1–4)	0.001
12 th hour	1 (0–1)	2 (1–3)	<0.001
24 th hour	1 (0–2)	1 (1–2)	0.258
VAS at cough			
1 st hour	3 (2–4)	5 (2–6)	0.024
2 nd hour	3 (1–3)	4 (2–5)	0.003
4 th hour	3 (1–3)	4 (2–5)	0.002
12 th hour	2 (2–3)	3 (2–4)	0.002
24 th hour	2 (1–3)	2 (2–3)	0.189

Continuous variables are expressed as the median (Q1: first quartile – Q3: third quartile). Continuous variables were compared with the Mann-Whitney U test. Statistically significant p-values are in bold. VAS: Visual analog scale.

Table 5. Comparison of the groups in terms of remifentanil requirement, morphine consumption, need for additional analgesia, and presence of nausea and vomiting

	Group-pre (n=30)	Group-post (n=30)	р
Intraoperative remifentanil requirement (mcg)	550 (425–720)	825 (600–1450)	0.001 ^β
Morphine consumption (mg)	13 (6–18)	28 (22–38)	< 0.001 ^β
Additional analgesic use, n (%)	4 (13.3)	17 (56.7)	< 0.001 ^δ
Nausea and vomiting, n (%)	1 (3.3)	6 (20.0)	0.103 ^ŏ

Continuous variables are expressed as the median β (Q1: first quartile – Q3: third quartile), and categorical variables are expressed as either frequency or percentage δ . Continuous variables were compared with a Student t-test or the Mann-Whitney U test, and categorical variables were compared using Pearson's chi-square test or Fisher's exact test. Statistically significant p-values are in bold.

stress response, have emerged. These applications have been shown to provide significant advantages in reducing complications and hospital costs.^[15]

These applications provide more effective results in minimally invasive techniques, such as VATS.^[9,16] ESPB, which is among the commonly used plane blocks, is an effective component of multimodal analgesia.^[8,16] However, there is still no clarity on the volume and concentration of local anesthetic used in ESPB block application, which is often preferred before surgical incision in the preoperative period. In addition, the mechanism of ESPB implementation remains controversial.^[17] Studies related to the application of ESPB in the postoperative period are also limited to rescue analgesia or rib fractures on a case-by-case basis.^[18,19] Studies in the literature generally compare the groups in which chest wall blocks were applied with the control groups in which chest wall blocks were not applied.^[9,20] Although chest wall

Table 4. Comparison of VAS scores between groups

blocks have been performed in the preoperative period in the literature, they can also be performed in the postoperative period in cases such as emergency surgery in clinical practice. In this study, the aim of our ESPB application in the postoperative period was to evaluate the preemptive effect of the preoperative ESPB application and to clarify whether the postoperative application will create a longer block.

Although the effectiveness of preemptive analgesia or block application is a controversial issue, it is still used in clinical practice. Preemptive analgesic administration is based on the limitation of this feature of the structures that carry the pain stimulus before the surgical incision. In this way, this method aims to reduce the pain that will occur and the stress response that may develop.^[4,9,21] In our study, intraoperative opioid consumption was more limited in patients in Group-Pre. Although VAS scores were low in the early postoperative period, morphine consumption was also low. This situation supports the preemptive application of ESPB. We also observed that the postoperative application of ESPB was not associated with a longer block time.

Uncontrolled surgical stress response is one of the essential factors in perioperative complications. This adversely affects patient outcomes in the intraoperative and postoperative periods. Therefore, opioids can be used to provide stable intraoperative anesthesia management. However, the adverse side effects of opioids limit this use.[20] Although there was no hemodynamic difference between the two groups in this study, the increase in intraoperative opioid consumption in Group-Post supports the hypothesis that opioids are used to provide stable hemodynamics. This suggests that preoperative administration of ESPB may limit the use of opioids and thus control the undesirable side effects of opioids and especially hyperalgesia that may develop due to remifentanil.

Limitations

This study has some limitations. Primarily, all block applications were performed by an anesthesiologist with approximately the same experience in both centers. Although this seems advantageous in terms of standardizing the block application and obtaining healthier results, it may not always be possible to apply it in clinical practice. Second, a nurse who blinded to the type of block questioned the patients regarding their pain levels, but intraoperative follow-up could not be performed by an anesthetist blinded to the study protocol. To eliminate this problem, we applied a standard anesthesia protocol to all patients. Third, postoperative pain monitoring was limited to only 24 hours. The purpose of this was to limit the follow-up period to 24 hours, because our goal was to examine the effectiveness of ESPB applications performed at different times in the early postoperative period. However, we continued to administer routine analgesic treatments to the patients after 24 hours. Finally, many biomarkers play a role in the stress response to surgery. However, these biomarkers could not be mentioned because the effects of the stress response on hemodynamics were among our study outcomes. Prospective randomized studies on these markers will provide guidance in this regard.

In conclusion, ESPB applied in the preoperative period is more effective in suppressing the surgical response in VATS applications, considering that it limits intraoperative opioid consumption and provides more effective analgesia in the postoperative period.

Disclosures

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Ethics Committee Approval: The study was approved by The Ankara City Hospital No. 1 Clinical Research Ethics Committee (Date: 06/04/2022, No: E1-22-2536).

Informed Consent: Written informed consent was obtained from all patients.

Peer-review: Externally peer-reviewed.

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Authorship Contributions: Concept – E.N.Z., M.Z., H.Y., A.A.; Design – E.N.Z., H.S., M.Z., H.Y., A.A.; Supervision – E.N.Z., H.S., S.Ş., M.Z., A.A.; Fundings – E.N.Z., S.Ş., H.Y.; Materials – E.N.Z., M.Z., H.Y.; Data collection &/or processing – E.N.Z., S.Ş., M.Z.; Analysis and/ or interpretation – E.N.Z., S.Ş., M.Z., H.Y.; Literature search – E.N.Z., M.Z., H.Y., A.A.; Writing – E.N.Z., H.S., S.Ş., M.Z., H.Y., A.A.; Critical review – E.N.Z., H.S., M.Z., A.A.

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