

# Evaluation of the effect of rectus sheath block and transversus abdominis plan block on sevoflurane and fentanyl consumption in laparoscopic cholecystectomy: A randomized controlled study

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## ABSTRACT

**Objectives:** We aimed to compare the total amount of inhalation anesthesia consumed and the amount of fentanyl used as an additional dose in cases where transversus abdominis plane and rectus sheath blocks were applied for postoperative analgesia.

**Methods:** Eighty patients aged 18-75 years, classified as ASA I-II, who were scheduled for laparoscopic cholecystectomy, were included. The patients were divided into two groups: those who received a combination of transversus abdominis and rectus sheath blocks under general anesthesia (Group B, n=40), and those who received analgesia with tramadol under general anesthesia (Group C, n=40). Throughout the procedure, both groups were monitored to maintain the entropy target value between 40-60 with a maximum MAC value of 1.3. At the end of the procedure, the total amount of inhalation agent consumed, and the additional need for superficial anesthesia/analgesia, as well as the total amount of fentanyl added as an extra dose, were recorded.

**Results:** Comparing the total amount of inhalation agent consumed throughout the case, it was found to be 27.05±7.43 mL in Group C (Control group) and 12.25±4.34 mL in Group B (Block group), with a statistically significant difference between the groups. There was a significant difference in the need for additional intraoperative and the total amount of fentanyl consumed between the groups (P<0.05).

**Conclusions:** In laparoscopic cholecystectomy cases, we observed that the use of transversus abdominis plane block and rectus sheath block combined with standardized general anesthesia monitored by entropy reduced the amount of inhalation agent consumed, as well as the need for additional intraoperative and total opioid consumption.

**Keywords:** Transversus abdominis plane block, rectus sheath block, entropy, sevoflurane, fentanyl

Inhalation anesthetics combined with opioid drugs are commonly used to prevent responses to pain but have side effects like prolonged recovery, nausea, vomiting, bowel dysfunction, respiratory depression, increased postoperative pain, environmental pollution, and higher healthcare costs. Therefore, min-

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imizing intraoperative anesthetic and analgesic use is important for perioperative physicians [1].

Electroencephalography (EEG) is widely recognized as one of the most objective methods for determining the depth of anesthesia [2]. In general anesthesia, changes in the EEG begin with induction, sedation, and maintenance [3-5]. Anesthesia depth neuromonitoring is based on the analysis of EEG changes resulting from the effect of anesthesia on cerebral blood flow and brain metabolism, and monitors that use it include the bispectral index (BIS), Narcotrend index, patient state index (PSI), entropy, SNAP index, and cerebral state index (CSI) [6].

Entropy provides two numerical values: state entropy (SE) and response entropy (RE), measured by using a low-impedance sensor on the frontal cortex EEG [7]. SE ranges from 0 (very deep anesthesia) to 91 (alertness), and RE ranges from 0 to 100, as displayed on the monitor [4]. SE (state entropy) reflects the cortical state more accurately and measures the hypnotic level. Instead of interpreting the RE (response entropy) value alone, it is more accurate to consider the difference between RE and SE, known as  $\Delta RE-SE$ , which reflects the frontal muscle EMG [8].

Peripheral blocks, aided by ultrasonography, have gained prominence in recent years as a crucial component of multimodal postoperative analgesia. These blocks effectively manage pain in the initial postoperative phase and help diminish the reliance on systemic opioids [9].

In this study, we aimed to compare the amount of inhalation and narcotic (fentanyl) used in anterior abdominal wall blocks applied for analgesia in laparoscopic cholecystectomy patients in whom anesthesia depth monitoring "entropy" monitor (target is in the 40-60 range) was used.

## METHODS

The research was started with the approval of the Tekirdağ Namık Kemal University Non-Interventional Clinical Research Ethics Committee on 26.07.2022 with the research protocol number 2022.138.07.05. The research period was planned as one year after ethics committee approval or until the specified number of cases was reached.

Eighty ASA I-II patients, aged between 18-75,

who were planned to undergo elective laparoscopic cholecystectomy, were included in the study by being divided into 2 groups using the randomization (sealed envelope method) method, whose written and verbal consent was obtained after being informed about the research protocol during the preoperative evaluation in the recovery unit. Group C (Control group) (n=40) was determined as Group B (Block group) (n=40), where analgesia was provided with tramadol during general anesthesia and analgesia was provided by adding transversus abdominis plane (TAP)+rectus sheath (RS) block to general anesthesia. The study, planned as randomized and single-blind, was conducted in a single center, in the operating room of Tekirdağ Namık Kemal University Hospital.

Sick sinus syndrome, kidney and liver failure, history of allergy to local anesthetics, intracranial vascular accident, patients with difficulty in establishing cooperation, chronic substance abuse, history of chronic opioid analgesia use, history of intraoperative awareness, pregnant women, morbidly obese patients, patients over 18 years of age. Patients who were younger, older than 75 years of age, diagnosed with a psychiatric disease, or had hemodynamic instability during the intraoperative period were not included in the study.

The same amount of correctly prepared drugs, in accordance with the literature, was administered to the patients in the preoperative, intraoperative and postoperative periods by the same experienced anesthesiologist, who did not monitor the patients during the study. The patients were monitored in the recovery unit for at least 20 minutes by another healthcare personnel who was not involved in the study. Those who were hemodynamically stable were sent to their services.

Demographic data (age, weight, height, body mass index) and ASA scores of the patients in both groups, who were taken to the operating room and monitored on the table in the supine position, were recorded. The patients were standardly monitored (ECG, SpO<sub>2</sub>, NBP). Vascular access was established with a 20-gauge intravenous cannula and 0.9% NaCl solution was provided. Entropy (GE Healthcare CARESCAPE Monitor B850, Entropy module) monitoring was added to both groups before induction as anesthesia depth monitoring. Two numerical entropy parameters were obtained on the monitor with the three electrodes of the entropy sensor placed in the frontotemporal region.

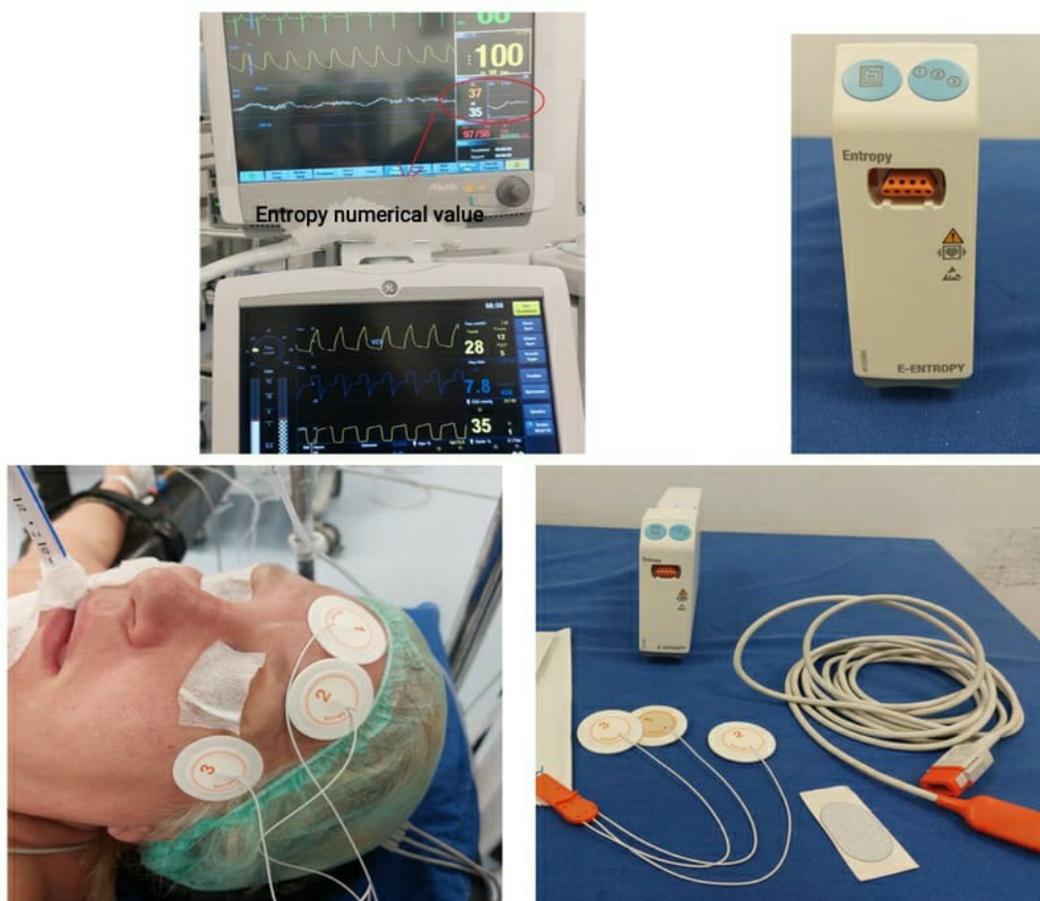
- SE-state entropy
- RE-response entropy (Fig. 1)

These values were recorded together with the initial entry hemodynamic parameters as baseline parameters. After 3 minutes of 100% preoxygenation, fentanyl (1-2 µg/kg), propofol (1-2.5 mg/kg), and rocuronium (0.6 mg/kg) were administered intravenously for standard induction of general anesthesia. After waiting for the neuromuscular blocker effect, all patients were orotracheally intubated with a laryngoscope. Anesthesia maintenance was provided with sevoflurane and an oxygen-air mixture of 50% O<sub>2</sub> and 50% air was used. Sevoflurane in anesthesia maintenance was started with the age-adjusted minimum alveolar concentration (MAC) value and the sevoflurane concentration was increased or decreased by evaluating the depth of anesthesia under the guidance of hemodynamics and entropy. The MAC was allowed to be increased up to a maximum of 1.3. 'Avance CS2 Anesthesia Delivery System' (GE Healthcare) was used as the anesthesia device in the study. For venti-

lation, the volume control mode (VCV) of the anesthesia device was selected with a tidal volume of 6-7 ml/kg, respiratory frequency 11-14/min, I: E ratio 1:2, and PEEP between 4-6.

For Group C (n=40), selected by randomization, patients were followed up under general anesthesia, and analgesia was administered with 100 mg tramadol intravenously after removal of the gallbladder. Group B (n=40) patients were intubated under general anesthesia and regional analgesia was performed with TAP (20 mL 0.25% bupivacaine) +RS (10 ml of 0.25% bupivacaine to coincide with the right trocar entry site) block under ultrasound guidance.

Sevoflurane concentration was adjusted to keep the entropy target value between 40-60 in both groups throughout the case, with a MAC value of maximum 1.3; starting, before and after induction, 1st minute (min.) after incision, pneumoperitoneum 5-10-15<sup>th</sup> min., 1<sup>st</sup> min. after pneumoperitoneum, post extubation, postoperative recovery 1<sup>st</sup> and 5<sup>th</sup> min.; The standard monitoring data of the cases, endtidal CO<sub>2</sub> values



**Fig. 1.** Monitoring screen, E-Entropy module, Entropy electrodes placement.

after orotracheal intubation, SE and RE entropy values, endtidal sevoflurane concentration (%) that kept the entropy value within the target range and simultaneous MAC values were measured and recorded at 5-minute intervals. Throughout the case, the need for analgesia/anesthesia status was monitored through hemodynamic parameters and clinical findings, along with the depth of anesthesia from the entropy monitor, hypertension (increase in the entrance mean arterial pressure by more than 20%), tachycardia (heart rate >120/min.), tears. In cases such as breathing, sweating, etc., or spontaneous breathing effort on the ventilator, an additional dose of fentanyl 1 µg/kg was administered intravenously and the total number of additional doses required was recorded. The maximum

fresh gas flow at the end of surgery was set to 4 l/min. Atropine 0.01 mg/kg and Neostigmine 0.03 mg/kg were administered intravenously to the patients to reverse the neuromuscular blockade effect. Patients whose spontaneous breathing was achieved were extubated. The patient, who remained under observation on the operating room table for another 5 minutes, was delivered to the recovery unit after the postoperative recovery 1<sup>st</sup> min. and the postoperative recovery 5<sup>th</sup> min. hemodynamic parameters were recorded.

In all patients included in the study, the total duration of surgery (min.), the pneumoperitoneum pressure applied during the case (mmHg), the total amount of fentanyl used (µg), the total amount of inhalation anesthetic consumed at the end of the case (mL) were noted.

**Table 1. Comparison of demographic information between groups**

	Group C (n=40)	Group B (n=40)	P value
<b>Age (year)</b>			
Mean±SD	46.65±13.99	51.75±12.65	0.547
Median (min-max)	47 (22-70)	53 (27-57)	
<b>Height (cm)</b>			
Mean±SD	1.64±0.08	1.62±0.07	0.569
Median (min-max)	1.67 (1.55-1.85)	1.64 (1.48-1.80)	
<b>Weight (kg)</b>			
Mean±SD	77.55±12.38	72.25 ±11.95	0.943
Median (min-max)	77.5 (57-98)	71.5 (48-95)	
<b>BMI (kg/m<sup>2</sup>)</b>			
Mean±SD	28.55±4.22	27.30±4.39	0.644
Median (min-max)	28.55 (17.20-36.20)	28.06 (17.80-36.90)	
<b>Gender (M/F), n (%)</b>			
Female	25 (62.5%)	29 (72.5%)	0.111
Male	15 (37.5%)	11 (27.5%)	
<b>ASA score, n (%)</b>			
ASA I	9 (22.5%)	14 (35%)	0.065
ASA II	31 (77.5%)	25 (65%)	
<b>Presence of Comorbidities, n (%)</b>			
(+)	24 (30%)	20 (25%)	0.064
(-)	16 (20%)	20 (25%)	

Data are shown as mean±standard deviation or median (minimum-maximum) or n (%) Group C=control group, Group b=Block group, BMI=body mass index, ASA=American Society of Anesthesiologists, M=male, F=female. Mann-Whitney U testi, Pearson Ki-square test, Fisher's Exact Testi  
P>0.05 statistically insignificant

## Statistical Analysis

SPSS Windows 25.0 (Statistical for social sciences for windows) statistical package program was used for data entry and statistical analysis. The study data were summarized with descriptive statistics (mean, median, standard deviation, minimum-maximum value, percentage, frequency). In the evaluation of the data, t-test, Mauchly's analysis of variance, Bonferroni correction analysis for repeated measures were applied. Chi-square test was used to compare categorical data. A value of  $P < 0.05$  was considered statistically significant.

## RESULTS

Our study included adult patients aged 18-75 years who were planned for ASA I and II laparoscopic cholecystectomy. We planned to compare the effect of transversus abdominis plan and rectus sheath block on sevoflurane and fentanyl consumption with entropy monitoring in these patients. For this purpose, the ages of the patients included in our study ranged between 22 and 70 years. When demographic data, ASA scores and comorbidities of the patients were compared between the groups, no statistically significant difference was found ( $P > 0.05$ ). Demographic data, ASA scores and patient comorbidity data of the study are shown in Table 1. (Table 1)

There was no statistically significant difference between the mean operation time and pneumoperi-

toneal pressure of the patients. ( $P > 0.05$ ) (Table 2) When the total amount of inhalation agent consumed during the total case duration was compared between the two groups, it was  $27.05 \pm 7.43$  ml in Group C and  $12.25 \pm 4.34$  mL in Group B, and a statistically significant difference was found between the groups ( $P < 0.05$ ) (Table 2).

The need for additional fentanyl doses during the intraoperative period was required in 30% of the cases. The need for additional fentanyl doses during the intraoperative period and the total amount of fentanyl consumed showed significant differences between the groups. Group C required more additional fentanyl doses intraoperatively than Group B (21 in Group C and 3 in Group B). In total,  $115 \pm 33.62$   $\mu$ cg and  $84 \pm 22.10$   $\mu$ cg fentanyl were consumed, respectively, and a statistically significant difference was found between the two groups ( $P < 0.05$ ) (Table 2).

According to our randomized controlled trial results, we demonstrated that preoperative TAP and rectus sheath block combined with general anesthesia reduced the intraoperative opioid and anesthetic agent consumption in laparoscopic cholecystectomy cases. We found that the amount of inhalation anesthetic agents consumed throughout the case, the need for intraoperative additional dose of fentanyl, and the total amount of fentanyl given throughout the case were significantly less in the laparoscopic cholecystectomy group (Group B) with TAP and RC block compared to the laparoscopic cholecystectomy group (Group C) without block ( $P = 0.002$ ,  $P < 0.001$ ,  $P = 0.001$ ).

**Table 2. Comparison of clinical information of patients in Group C and Group B**

	Group C (n=40)	Group B (n=40)	P value
<b>Total duration of surgery (minute)</b>	40 (30-50)	40 (30-50)	0.213
<b>Pneumoperitoneum pressure (mmHg)</b>	14 (12-16)	15 (12-18)	0.386
<b>Amount of inhalation anesthetic consumed (mL)</b>	33.5 (19-48)	13.5 (6-21)	<b>0.002*</b>
<b>Intraoperative need for additional dose of fentanyl</b>			
(+)	21 (52.5%)	3 (7.5%)	<b>&lt;0.001*</b>
(-)	19 (47.5%)	37 (92.5%)	
<b>Total amount of fentanyl (<math>\mu</math>cg)</b>	125 (60-190)	95 (55-135)	<b>0.001*</b>

Data are shown as mean  $\pm$  standard deviation or n (%). Group C=control group, Group B=block group.

Mann-Whitney U testi, Student t test

\* $P < 0.05$  statistically significant

**Table 1. Comparison of SE values of Group C and Group B**

State entropy (SE)	Group C (n=40)	Group B (n=40)	P value
Initial	89.18±3.28	88.30±3.08	0.724
Before induction	87.38±4.55	88.18±2.77	<b>0.042*</b>
After induction	44.75±5.05	45.18±5.88	0.441
After incision 1 <sup>st</sup> min	49.45±6.60	51.13±6.73	0.978
Pnömooperitoneum 5 <sup>th</sup> min	45.75±5.53	47.28±5.89	0.590
Pnömooperitoneum 10 <sup>th</sup> min	48.35±5.87	48.10±5.62	0.946
Pnömooperitoneum 15 <sup>th</sup> min	48.48±5.93	48.93±5.86	0.740
After pneumoperitoneum 1 <sup>st</sup> min	50.18±8.21	53.20±6.34	0.840
After extubation	82.78±7.45	82.50±8.25	0.838
Postoperative recovery 1 <sup>st</sup> min	86.43±5.25	87.33±2.95	0.005
Postoperative recovery 5 <sup>th</sup> min	89.30±3.04	89.43±2.71	0.381

Data are shown as mean±standard deviation. Group C=control group, Group B=block group, SE=state entropy, min=minute.

Student t test, Paired Samples t test

\*P<0.05 statistically significant

The pre-induction SE (static entropy, entropy of state) values of Group B were statistically significantly higher than Group C (P=0.042, P<0.05). When SE measurements were compared between the groups, there was no statistically significant difference (P>0.05) between baseline, post-induction, 1<sup>st</sup> min. after incision, 5<sup>th</sup> min., 10<sup>th</sup> min. and 15<sup>th</sup> min. after pneumoperitoneum, 1<sup>st</sup> min after pneumoperitoneum,

1<sup>st</sup> min., and 5<sup>th</sup> min. after postoperative recovery (Table 3).

When RE (respond entropy) measurements were compared between the groups, there was no statistically significant difference (P>0.05) between baseline, pre-induction and post-induction, 1<sup>st</sup> min. after incision, 5<sup>th</sup> min., 10<sup>th</sup> min. and 15<sup>th</sup> min. after pneumoperitoneum, 1<sup>st</sup> min. after pneumoperitoneum, 1<sup>st</sup>

**Table 2. Comparison of RE values of Group C and Group B**

Respond entropy (RE)	Group C (n=40)	Group B (n=40)	P value
Initial	96.88±1.84	97.00±2.23	0.383
Before induction	95.53±3.81	96.40±2.83	0.718
After induction	46.95±5.19	47.80±6.82	0.053
After incision 1 <sup>st</sup> min	51.85±6.68	53.00±6.52	0.513
Pnömooperitoneum 5 <sup>th</sup> min	48.75±5.75	49.05±5.78	0.874
Pnömooperitoneum 10 <sup>th</sup> min	50.78±6.20	49.90±6.08	0.573
Pnömooperitoneum 15 <sup>th</sup> min	50.40±5.67	50.68±5.80	0.921
After pneumoperitoneum 1 <sup>st</sup> min	52.50±8.32	54.40±7.46	0.857
After extubation	91.65±6.58	91.75±7.68	0.380
Postoperative recovery 1 <sup>st</sup> min	94.65±4.17	95.78±3.04	0.612
Postoperative recovery 5 <sup>th</sup> min	96.65±2.15	97.23±1.96	0.315

Data are shown as mean±standard deviation. Group C=control group, Group B=block group, RE=respond entropy, min=minute.

Student t test, Paired Samples t test

min. after extubation, 1st min. and 5th min. after post-operative recovery (Table 4).

Since there should be no difference between the entropy values to make a statistical comparison between the groups, we were able to achieve this value with the statistical results in Tables 3 and 4.

## DISCUSSION

The commonly preferred combination of anesthesia in laparoscopic cholecystectomy cases is induction of general anesthesia followed by maintenance with a combination of inhalation agents and opioids. However, these agents have undesirable effects such as prolonging recovery time, causing postoperative nausea and vomiting, causing intestinal dysfunction and respiratory depression, causing postoperative rebound pain, increasing environmental pollution and increasing health costs [10, 11]. Therefore, minimizing intraoperative consumption of anesthetic and analgesic agents should be a priority for every anesthesiologist [12].

Fascial plane blocks performed after intubation for preemptive analgesia to minimize the trauma of the surgical incision and to reduce the incidence of postoperative chronic pain have become popular [13-15]. TAP and rectus sheath block is a peripheral block method that blocks somatic nerves in the anterior abdominal wall. Bilateral TAP and rectus sheath block has been successfully used for pain control in laparoscopic cholecystectomies [16, 17].

There are a limited number of studies in the literature on intraoperative anesthetic agent consumption in laparoscopic surgeries combining TAP and rectus sheath block with general anesthesia [18, 19]. Our study aimed to evaluate the effect of TAP and RC block on inhalation agent and opioid consumption. In our study, we found that sevoflurane consumption was less in the Group B compared to the Group K at the same depth of anesthesia. At this point, we believe that TAP and rectus sheath block contribute to multimodal analgesia strategies by reducing inhaled gas consumption.

Kokulu *et al.* [19] compared the cost and anesthetic agent consumption in cases with and without TAP block, with intraoperative Target BIS kept at 40-50 in laparoscopic cholecystectomy surgery. They reported that anesthetic agent use decreased in the TAP block group. They did not find a statistical difference

in the amount of intraoperative opioid consumption between the two groups. Contrary to the results of this study, sevoflurane consumption was lower in laparoscopic cholecystectomy cases where we applied TAP and rectus sheath block in our study. In addition, the need for intraoperative additional fentanyl dose and the total amount of fentanyl consumed were found to be lower in Group B. We think that this is due to the preemptive fascial plane blocks. Monitoring the depth of anesthesia with entropy gave us different results.

Karaman *et al.* [1] studied the intraoperative remifentanyl and sevoflurane consumption in the group in which TAP block was combined with patients undergoing total abdominal hysterectomy (TAH) under general anesthesia. They found that total remifentanyl and sevoflurane consumption was significantly lower in the group in which TAP block was added. They found that QoR-40 (Quality of Recovery score) was significantly higher in Group TAP patients. They reported that combining TAP block with general anesthesia reduced opioid and anesthetic consumption in TAH cases and provided a better postoperative period. In our study, the total amount of inhalational anesthetic consumed during the case was statistically significantly lower in Group B than in Group K. Our study also obtained similar results with this study.

Bhattacharjee *et al.* [20] investigated the effect of TAP block and saline infusion after general anesthesia on intraoperative hemodynamic parameters and intraoperative fentanyl consumption in patients undergoing TAH surgery. They found that the heart rate, systolic blood pressure and intraoperative fentanyl requirement during surgery were significantly higher in the saline group compared to the block group. In conclusion, they showed that TAP block combined with general anesthesia before incision reduced intraoperative fentanyl requirement, prevented hemodynamic responses to surgical stimuli and provided appropriate postoperative analgesia. Our study was parallel to this study and the amount of intraoperative additional fentanyl dose administered was statistically significantly higher in Group K compared to Group B.

Opioid agents contribute to postoperative pain by inhibiting nociception. However, due to undesirable side effects, different methods and various drugs are used for postoperative pain management instead of opioids [21]. Regional regional regional anesthesia is a very popular method in this sense. In our study, we

used a combination of two facial plan blocks for post-operative analgesia.

Insufficient depth of anesthesia may not suppress somatic and autonomic reflexes sufficiently and may be harmful especially in patients with limited cardiac reserve. Excessive depth of anesthesia may also suppress vital functions and may cause complications such as coma and death [3]. To prevent anesthesia complications especially in high-risk patient groups, anesthesia depth measurement should be added to standard monitoring [22]. In our study, we used entropy monitoring, which is considered more recent, to monitor the depth of anesthesia.

The importance of environmentally sustainable health services has also become more on the agenda [23]. The provision of various health services, such as operating rooms, consumables, waste volumes, high energy systems, anesthesia gas systems, etc. causes a considerable amount of greenhouse gas emissions [24]. In the study examining the effect of anesthesia types on carbon footprint, McGain *et al.* [25] mentioned that general, regional and combined anesthetics have similar effects in terms of CO<sub>2</sub> equivalence in greenhouse gas emissions. However, of the 3 main factors affecting the carbon footprint (disposable equipment, electricity consumption of devices, and drugs used), it is stated that the biggest change can be made through drugs. It has been shown that sevoflurane, one of the inhalation anesthetic agents, occupies a significant place in greenhouse gas emissions, especially in patients undergoing general and combined anesthesia. In a study by Struys *et al.* [26] on the environmental effects of anesthesia, it was reported that inhalation anesthetics such as nitrogen oxide and halogenated ethers have a serious global warming potential because they are important greenhouse gases. As a result, inhalation anesthetics consumed during the operation were found to cause more than 50% of the peroperative greenhouse gases. Therefore, reducing the amount of inhalation anesthetic agents consumed by measuring the depth of anesthesia is an ethical obligation for every anesthesiologist in these days of global climate change.

One of the Principles of Environmentally Sustainable Anesthesia in the Global Consensus Statement of the World Federation of Societies of Anesthesiologists aims to reduce atmospheric waste of volatile agents to reduce the carbon footprint [27]. However, in the face

of ever-increasing health expenditures, cost control is also important. It should be aimed to identify various strategies to reduce the use of inhalation anesthetic agents due to their high costs [28].

### Limitations

Our study was conducted at a single center; conducting multicenter randomized controlled trials with larger numbers of patients under similar standards would raise awareness for reducing consumption of inhalation anesthetic agents.

### CONCLUSION

The strengths of our study include the use of the entropy monitor during the operation, the fact that the TAP and rectus sheath block were performed by an experienced anesthesiologist and similar block success was achieved in each block application (we visually monitored the distribution of local anesthesia in the target area simultaneously with ultrasonography and confirmed that the blocks were performed effectively), the same physician performed the data collection phase, and we standardized anesthesia for both groups with patient group selection factors. The weaknesses of our study are that the operations were performed by different surgeons even though the same surgical technique was applied, dermatomal sensory block levels could not be determined after TAP and rectus sheath block, and postoperative analgesia follow-up was not performed in every patient. As a result, we have shown that monitoring the depth of anesthesia of patients undergoing preoperative block may support cost and environmental protection, in addition to less drug use and fewer side effects.

### Ethical Statement

This study was approved by the Tekirdağ Namık Kemal University Non-Interventional Clinical Research Ethics Committee on 26.07.2022 with the research protocol number 2022.138.07.05.

### Authors' Contribution

Study Conception: EK, CA; Study Design: EK, AG; Supervision: EK, IY; Funding: EK, IY; Materials: EK, AG; Data Collection and/or Processing: EK, BT; Statistical Analysis and/or Data Interpretation: EK,

BT; Literature Review: EK, AG; Manuscript Preparation: EK, IY and Critical Review: EK, CA.

### Conflict of interest

The authors disclosed no conflict of interest during the preparation or publication of this manuscript.

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### Editor's note

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