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Dynamic thiol/disulphide balance in patients undergoing hypotensive anesthesia in elective septoplasties

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Abstract

Objective: We aimed to investigate the effects of hypotensive anaesthesia on oxidative stress with serum thiol/disulphide balance in patients undergoing elective septoplasty procedures under general anaesthesia.

Methods: Seventy-two patients between the ages of 18-60, with a physical condition I –II, according to the American Society of Anesthesiologists, were included in this prospective observational study. Septoplasty was chosen for standard surgical stress. According to the maintenance of anaesthesia, patients were divided into the groups as Hypotensive Anaesthesia (n = 40) and Normotensive Anaesthesia (n = 32). Serum thiol/disulphide levels were measured by the method developed by Erel & Neşelioğlu.

Results: The native thiol and total thiol values of both groups measured at the 60th min intraoperatively were significantly lower than the preoperative values (both $P < .01$). Intraoperatively, at the 60th min, there was no significant difference in terms of post-native thiol and post-total thiol levels between hypotensive and normotensive anaesthesia groups ($P = .68$ and .81, respectively). Age >40 years and female gender were found to have a significant effect on dynamic oxidative stress ($P = .002$ and .001, respectively).

Conclusion: This pilot study has found that hypotensive anaesthesia had no adverse effect on dynamic thiol/disulphide balance in elective surgeries.

1 | INTRODUCTION

Hypotensive anaesthesia is widely used in head and neck surgery.[1-4] The mean arterial blood pressure is lowered by 20%-30% with various drugs and methods to alleviate blood loss or to improve the quality of the surgical field.[5,6] There are concerns about tissue ischemia with controlled hypotension due to hypoperfusion of organs.[7] Due to differences in patients' susceptibility to organ hypoperfusion; it is not clear to what extent blood pressure may be reduced. Serum lactate level, base deficit, and infrared spectroscopy have been used to monitor the adequacy of organ blood flow.[8,9] Another suggested method to monitor tissue ischemia is to measure oxidative stress level.[10]

Oxidative stress is highly susceptible to patient-and procedure-related factors like smoking, drugs administered in the perioperative period, general anaesthesia, and surgical trauma.[11-13] Erel and Neşelioğlu developed an automated method that directly measures serum thiol/disulphide levels.[14] Thiols are organic compounds that protect against cell damage caused by reactive oxygen species.[15,16] The thiol/disulphide balance may be used as a marker of oxidative stress level, thus tissue ischemia.

[Correction added on 23 September 2021, after first online publication: Affiliations were corrected for the third, and fifth to seventh authors in this version.]

We designed a prospective, observational study to investigate the effect of hypotensive anaesthesia on oxidative stress with serum thiol/disulphide balance in elective septoplasty operations.

What is already known

- Oxidative stress is highly susceptible to patient-and procedure-related factors like smoking, drugs administered in the perioperative period, general anaesthesia, and surgical trauma.
- Erel and Neşelioğlu developed an automated method that directly measures serum thiol/disulphide levels.
- The thiol/disulphide balance may be used as a marker of oxidative stress level, thus tissue ischemia.

What this paper adds

- Hypotensive anaesthesia had no negative effect on dynamic thiol/disulphide balance in patients undergoing elective surgeries.

2 | MATERIAL AND METHODS

This study was designed as a prospective observational study. The study protocol was recorded in the Clinical Trials database (NCT03501563). Ethics committee approval was obtained from Recep Tayyip Erdogan University Research Ethics Committee (Number: 2018/63) and the study was performed in conformance with the ethical guidelines of the Declaration of Helsinki. All patients were informed in detail about the objective of the study and signed informed written consent forms.

Patients scheduled for elective septoplasty surgery due to nasal septum deviation between April-September 2018, aged 18-60 years with American Society of Anesthesiologists physical status of 1-2 were included in the study. All surgeries were performed by the same surgical team.

Patients with uncontrolled hypertension, diabetes mellitus, cerebrovascular disease, morbid obesity defined as body mass index ≥35, anaemia, pregnancy, chronic kidney disease, carotid artery stenosis, history of antioxidant drug use during the last month, and with known allergies to study drugs were excluded.

2.1 | Control and study groups

Patients' anaesthetic charts were reviewed independently by two anaesthesiologists. Patients were allocated to normotensive anaesthesia group (Group N) if their mean arterial blood pressures were maintained within ±20% of preoperative mean arterial blood pressure. Patients were allocated to the hypotensive anaesthesia group (Group H) if their mean arterial blood pressures were maintained between 55 and 65 mmHg, or patients' systolic blood pressures were maintained as <100 mmHg.

2.2 | Anaesthesia management

Perioperative anaesthetic care was standardized as follows: 5 mL/kg of iv isotonic fluid was infused within 30 min before the induction of anaesthesia, through an 18-gauge cannula inserted into the left brachial vein. A 20-gauge iv cannula inserted into the dorsum of the right hand was used to draw venous blood samples. Three-lead electrocardiogram, noninvasive arterial blood pressure on right arm, peripheral oxygen saturation(SpO2) via left hand, end-tidal carbon dioxide ($EtCO_2$), body temperature on the left axilla, neuromuscular functions (Datex-Ohmeda M-NMT module; Datex-Ohmeda), bispectral index (BIS, Vista Monitoring System) were monitorised.

Blood pressure was measured on the operating table, when the infusion of isotonic fluid finished, and patients rested for 5 min in supine position. Baseline mean arterial blood pressure was calculated by averaging three consecutive measurements at 5 min intervals.

During preoxygenation, 1 μg/kg iv remifentanil was infused within 60 seconds, and remifentanil infusion was started at a rate of 0.25-0.5 μg /kg-/ min. Neuromuscular blockage was obtained with 0.6 mg/ kg rocuronium bromide. Train of four test (TOF) was used which was performed by peripheral nerve stimulator with the purpose to determine the degree of muscle relaxation. Orotracheal intubation was performed after the disappearance of twitches in TOF. Mechanical ventilation was induced with 6 ml/kg tidal volume by ideal body weight. 5 cmH_2O positive end-expiratory pressure with volume-controlled mode (Dräger Primus anaesthesia machine, Dräger Medical, Lübeck, Germany). Anaesthesia was maintained with 3%-6% desflurane, adjusted to maintain a BIS value within 50%-60% with 2/ L/min fresh gas flow (50% O_2). Any TOF response of >2 twitches were treated with 0.2/mg/kg rocuronium bromide.

Hypotension was defined as mean arterial blood pressure (MAP) <50 mmHg and treated with 5 mg of iv ephedrine. Bradycardia was defined as heart rate (HR) <45 beats /min and treated with 0.5 mg of iv atropine.

All patients received an infusion of 100 mg of iv tramadol and a bolus dose of 8 mg of iv ondansetron 15 min before end of the surgery. After completion of surgery, and following a TOF value of >75%, 15 μg/kg iv atropine and 50μg/kg iv neostigmine were administered. Following a BIS value >80%, and patient's compliance to verbal commands, trachea was extubated. All patients were observed in the postoperative recovery unit.

2.3 | Data collection

MAP, HR, SpO_2, $EtCO_2$, BIS values were recorded before anaesthesia induction, after tracheal intubation, and at 5 min intervals thereafter. The venous blood samples were taken 10 min before the

preoperative anaesthesia induction and at the 60th min intraoperatively. All blood samples and MAP, HR, SpO2, EtCO2, BIS values were collected and read by the same researcher.

2.4 | Biochemical analysis

Venous blood samples were collected in biochemistry tubes containing coded anticoagulants and serum separators. Tubes were centrifuged at 1500 rpm for 10 min and stored in −80°C dry environment by freezing. Thiol/Disulphide homeostasis tests were performed using Erel and Neselioğlu's spectrophotometric method.[14] Disulphide bonds were first reduced to form free functional thiol groups with sodium borohydride. Accumulating sodiumborohydride was consumed and removed with formaldehyde to prevent reduction of 5,5′-dithiobis- (2-nitrobenzoic) acid (DTNB). After reaction with DTNB, all thiol groups were reduced, including "disulphide", "native thiol" and "total thiol" groups. After determination of native and total thiols, disulphide amounts were calculated as disulphide/native thiol, disulphide/total thiol, and native thiol/total thiol percentage rates.

2.5 | Statistical analysis

Sample size was calculated as 31 for each group with G*Power software (Franz Faul, UniversitätKiel) version 3.1.9.4 (effect size: 0.65, type 1 error:0.2, type 2 error: 0.05).

Statistical analysis was done with SPSS for Windows version 22 (IBM). Distribution of continuous variables was tested with the Kolmogorov-Smirnov test. Normally distributed data were given as mean ± standard deviation. Others were given as median (interquartile range). Categorical data were given as number (%). Intergroup differences as for age, gender, ASA, body mass index (BMI), surgical time, baseline physiologic values and laboratory measurements were analysed with dependent samples t-test.

Repeated measurements (MAP, HR, SpO_2, EtCO2, BIS values, laboratory measurements) were examined with Analysis of Variance test. Due to low sample size, the effects of patient characteristics (age, gender, BMI) on thiol/disulphide levels were examined with binomial test. Due to lack of any significant correlation, further analysis was not performed. A P value < .05 was considered statistically significant.

3 | RESULTS

Study flow diagram is given in Figure 1. Briefly, a total of 92 patients were included in the study; data from 72 patients were analysed. Patient characteristics are given in Table 1.

Native thiol, total thiol values and native thiol/total thiol ratio are given in Figure 2. Preoperative native thiol (Group H: 402.7 ± 32.2 lmol/L, Group N: 406.7 ± 33, 4 lmol/L) and total thiol values (Group H: 444.4 ± 39.5 lmol/L, N: 444.4 ± 35.9 lmol/L) were similar between hypotensive and normotensive anaesthesia groups (P = .61, and .96, respectively).

Post- native thiol (Group H: 324.5 ± 56.5 lmol/L, Group N: 328.3 ± 28.2 lmol/L)and post- total thiol values (Group H: 360.4 ± 59.9 lmol/L, Group N: 363.1 ± 35.4 lmol/L) measured at the 60th min intraoperatively were significantly lower than the preoperative values (P < .01 and .01, respectively). However, preoperative native thiol/total thiol ratio was preserved at the 60th min in hypotensive and normotensive anaesthesia groups (P > .05).

Intraoperatively, at the 60th min, there was no significant difference between hypotensive and normotensive anaesthesia groups in terms of post- native thiol and post- total thiol levels (P =.68 and .81, respectively) (Figure 2).

Disulphide levels are given in Figure 3. Serum post-disulphide levels, disulphide/native thiol ratios and disulphide/total thiol ratios at the 60th min were similar to the preoperative values in both groups (P > .05).

The effects of age, gender, ASA, BMI, and surgical time on dynamic oxidative stress measurements were evaluated by logistic regression analysis. Age >40 years and female gender were found to have a significant effect on oxidative stress levels (P = .002 and .001, respectively).

4 | DISCUSSION

This study has demonstrated that hypotensive anaesthesia is a safe method in terms of oxidative stress in patients with ASA 1- 2 status. Serum thiol and disulphide measurements decreased to normal values in all patients who underwent septoplasty operation under general anaesthesia. However, there was no significant difference between hypotensive and normotensive anaesthesia groups in terms of post- native thiol and post- total thiol levels and hemodynamic management had no observable effect.

Septoplasty procedure was chosen as it has minimal and standard surgical stress. Also, there are previous studies investigating oxidative stress in septoplasty procedures, providing a rich database to compare our results.[17,18] However, we are not aware of any study investigating the effects of hypotensive anaesthesia on oxidative stress.

It has been reported that ischemic damage due to hypoperfusion may develop in some surgeries where hypotensive anaesthesia is applied.[19-22] Some studies have used organ-specific biomarkers[23,24] to demonstrate ischemia at the cellular level, while others have used oxidative stress markers.[25]

We did not expect ischemia at the organ level because ASA I and II patients without diabetes and similar chronic diseases were included in our study. Therefore, oxidative stress markers were preferred to detect ischemia at the cellular level. The reduction of thiol levels at the 60th min in both groups is consistent with the literature.[26-29] The similar level of decline suggests that the cause is due to surgical stress or anaesthetic drugs rather than hemodynamic management.

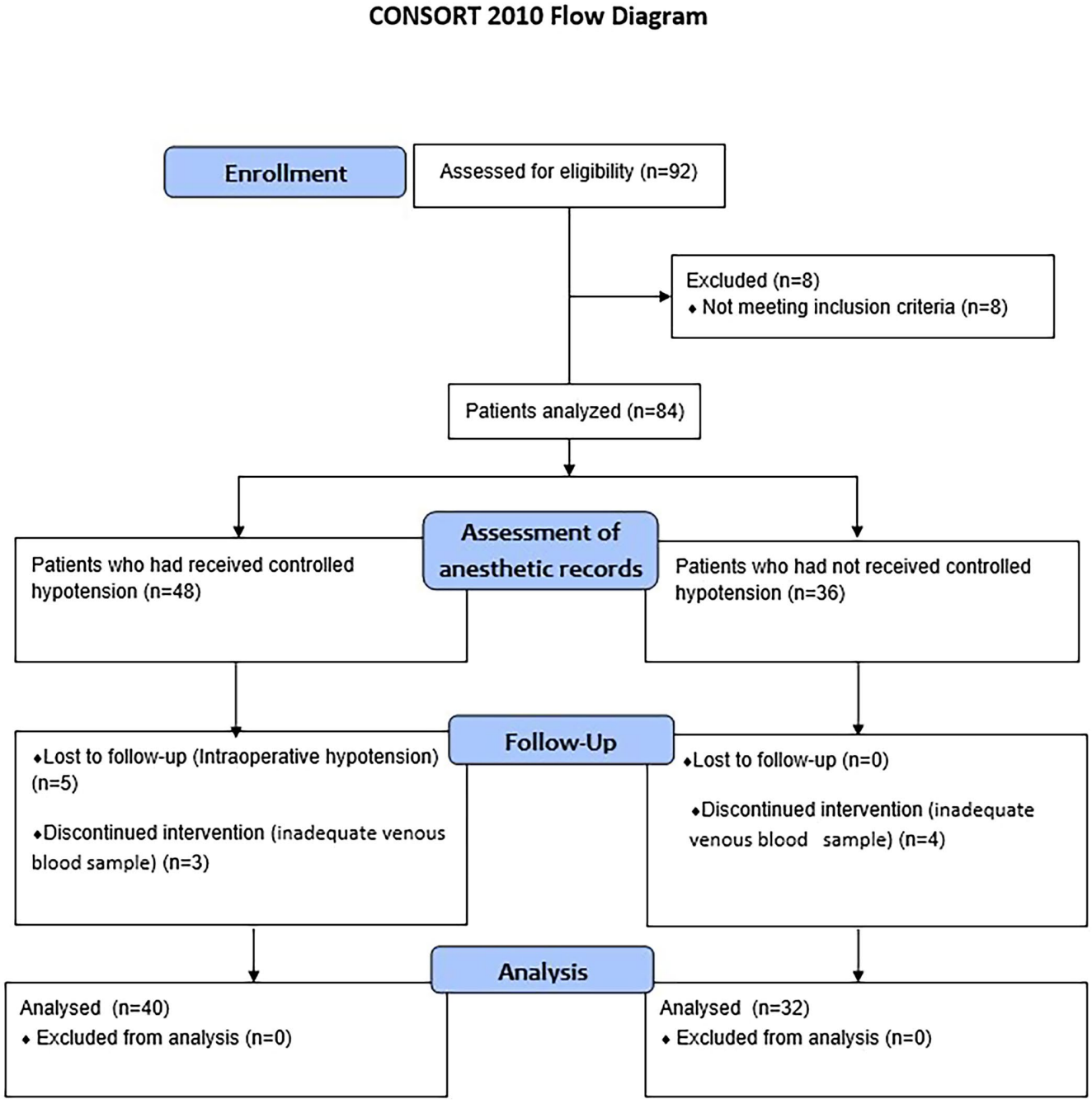

FIGURE 1 Study flow diagram

The effects of surgical stress are manifested primarily by surgical trauma and then by stress response defined by the host's neurohumoral, immunological and metabolic changes.[30] The main mechanism is increased catabolism and oxygen consumption. In this study, surgical stress was standardized by selecting a uniform operation and performing the procedure by the same surgeons. The increase in catabolism and oxygen consumption was standardized with anaesthesia applied under the guidance of neuromonitorisation and neuromuscular monitoring.

Akın et al evaluated the effect of general and spinal anaesthesia on oxidative stress parameters in ASA I-II patients undergoing elective caesarean delivery and reported that general anaesthesia had more negative effects on dynamic thiol disulphide balance.[31] This result suggests that general anaesthesia may be effective in decreasing thiol levels in our study.

Çukurova et al examined oxidative chromosomal damage in bronchoalveolar lavage samples and in plasma in lumbar discectomy surgeries performed with sevoflurane and desflurane. The authors noted that both inhalation agents cause damage in bronchoalveolar cells. They also reported that local genotoxicity and systemic oxidized chromosomal damage were similar in both groups.[32]

TABLE 1 Patient characteristics. Values are expressed as mean ± standard deviation, or number (percent %)

	Group H (n = 40)	Group N (n = 32)	*P* value
Age, years	31 ± 11.4	30.8 ± 9.3	.957
Female gender, n (%)	24 (%60)	10 (%31)	.677
BMI, kg m^{-2}	24.4 ± 4.6	24.7 ± 3.8	.698
ASA score I, n (%)	36 (%90)	21 (%65)	.539
Duration of operation, min	83.2 ± 14.7	83.3 ± 11.9	.992
Mean arterial blood pressure, mmHg			
Basal	92.8 ± 9.6	95.4 ± 9.7	.248
Post-induction	68.2 ± 5.5	78.6 ± 6.4	**<.001**
15 min	62.7 ± 4.8	76.8 ± 5.1	**<.001**
30 min	60.7 ± 3.4	74.2 ± 5.2	**<.001**
60 min	60.9 ± 2.8	75.3 ± 5.1	**<.001**

Abbreviations: BMI, Body Mass Index; Group H, Hypotensive Anaesthesia; Group N, Normotensive Anaesthesia.

thiol, total thiol, native/total thiol ratios; and higher disulphide, disulphide/native thiol, disulphide/total thiol ratios in patients with masked hypertension.[34] Since their results are contradictory to ours, we believe that our results are not affected by masked hypertension. Although anaesthesia and surgical methods are standardized, it is possible that phenotypic differences that have not been detected yet have affected the outcome.[35]

Another limitation of our study is that patients were not randomized in terms of smoking, diet and similar lifestyle parameters that might affect oxidative stress. Since the number of patients in this pilot study is insufficient to examine the effects of these and other parameters on oxidative stress, randomized controlled trials involving a large number of patients are needed.

In addition, the variable course of MAP in our normotensive group resulted in similar oxidative stress levels between the groups. Therefore, a controlled study in which normotensive anaesthesia is standardized could provide more reliable results. Patients with uncontrolled hypertension were excluded from this study, and patients with ASA I-II status were included. However, there are studies where only ASA I patients were analysed.[2] It is possible that including different ASA groups may provide different results.

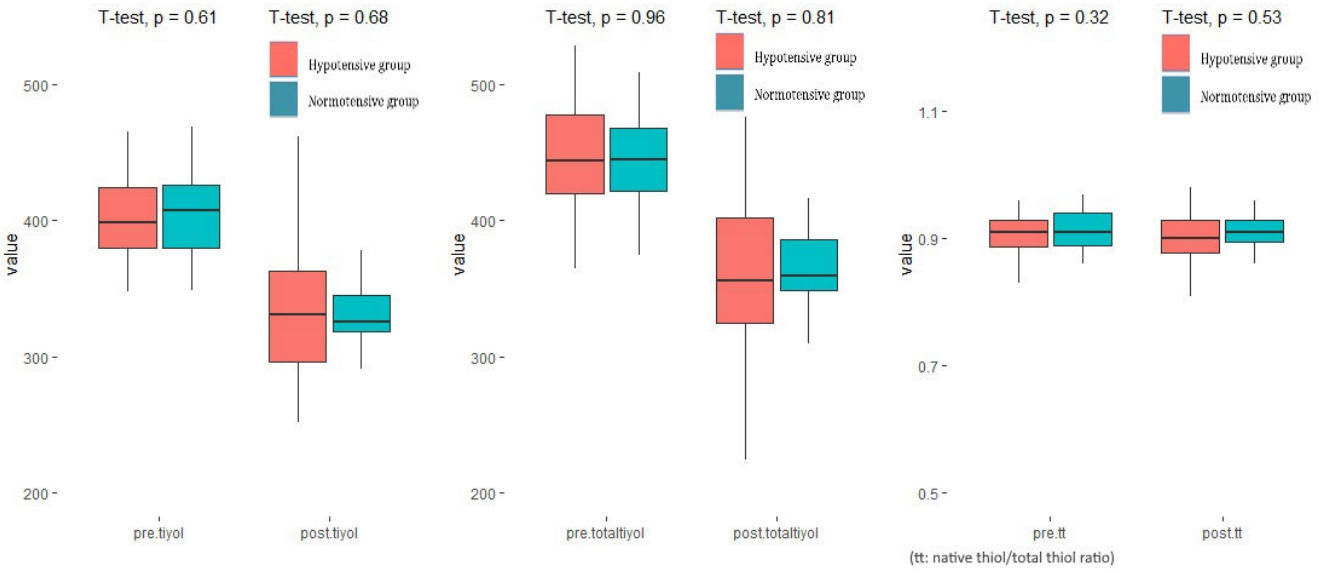

FIGURE 2 Native thiol, total thiol values and native thiol/total thiol ratio. Pre, Preoperative; Post, Intraoperatively at the 60th min; tt, native thiol/total thiol ratio ■ Hypotensive group; ■ Normotensive group

Özcan et al showed that the effects of sevoflurane on oxidative damage were more pronounced in their studies comparing to the effects of sevoflurane and desflurane on thiol-disulphide homeostasis in patients undergoing laparoscopic cholecystectomy.[33]

Çukurova and Özcan's results suggest that inhalation anaesthetic agents may cause varying levels of oxidative damage in different individuals.[32,33] Therefore, the adaptability of the results of our study to different patient populations or anaesthetic agents is limited.

Patients whose blood pressure cannot be reduced to hypotensive targets may possibly have masked hypertension. Ateş et al studied the effect of masked hypertension (office measurements >130/80 mmHg or at home measurements >135/85 mmHg) on dynamic thiol/disulphide balance. They reported lower native

On the other hand, we could include another oxidative stress marker to compare our results and this would increase the strength of our study. Furthermore, patient-related factors known to affect susceptibility to oxidative stress such as smoking and perioperative medications could not be assessed.

Lastly, this pilot study aimed to investigate the effects of hypotensive anaesthesia alone. In this regard, a surgery such as septoplasty was chosen, where bleeding and morbidity are not expected. Repeating this study using more invasive surgical procedure may yield different results.

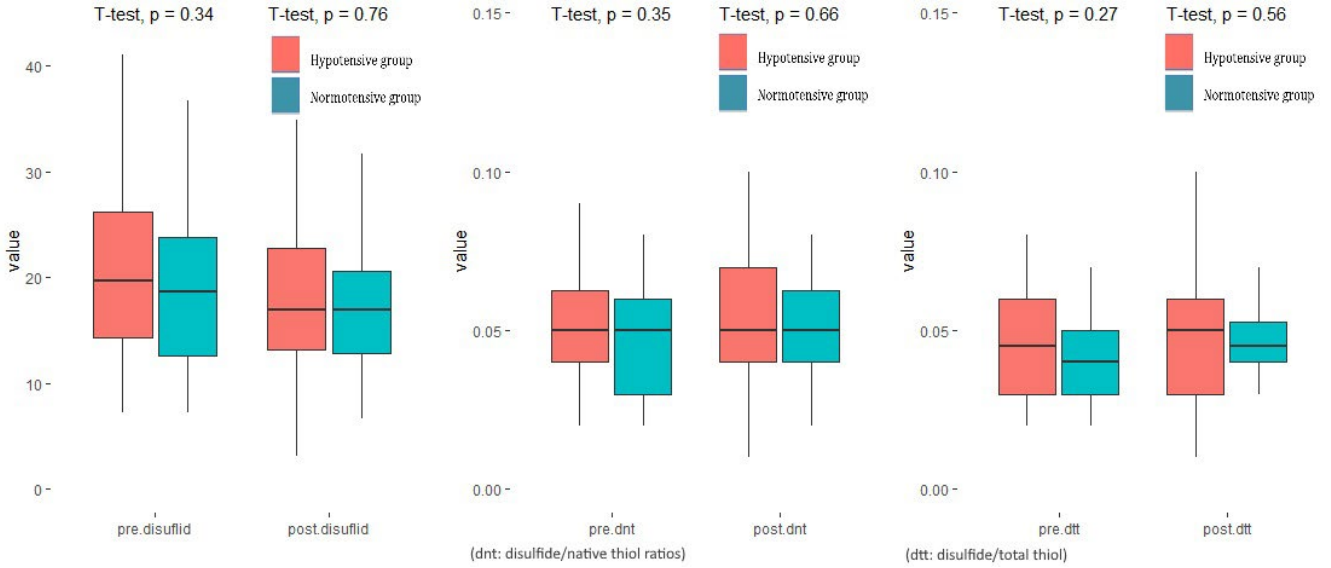

FIGURE 3 Disulphide levels. Dnt, disulphide/native thiol ratios; dtt, disulphide/total thiol

In conclusion, it was found that hypotensive anaesthesia had no negative effect on dynamic thiol/disulphide balance in patients undergoing elective surgeries with a physical condition I –II in this pilot study. Other controlled clinical studies randomizing lifestyle parameters and examining dynamic thiol/disulphide homeostasis in more invasive surgeries are required.

CONFLICT OF INTEREST

The authors declare no conflict of interest to disclose.

DATA AVAILABILITY STATEMENT

All data generated or analysed during this study are included in this published article.

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