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EFFICACY OF SEVOFLURANE VERSUS PROPOFOL FOR INTUBATION WITHOUT USING NEUROMUSCULAR BLOCKERS IN CHILDREN UNDERGOING ELECTIVE SURGERY AT TERTIARY CARE HOSPITAL.

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ABSTRACT:

BACKGROUND: The process of pediatric endotracheal intubation in the context of elective procedures presents distinct difficulties, as the selection of induction agents directly influences the resulting clinical results. Using neuromuscular blockers has inherent hazards, necessitating investigating other approaches to attain ideal intubation circumstances. The present research aimed to examine and compare the effectiveness of sevoflurane and propofol in pediatric intubation, particularly in the absence of neuromuscular blockers. OBJECTIVE: To compare the efficacy of Sevoflurane versus Propofol for intubation without using neuromuscular blockers in children undergoing elective surgeries at a tertiary care hospital. METHODS: A randomized controlled trial was conducted in the Department of Anesthesiology, Patel Hospital Karachi, over a period of six months. The research comprised a cohort of sixty-two pediatric patients aged 1 month to 5 years. These patients were categorized as ASA status I-II. The individuals in question were slated to undergo elective medical operations while under the effects of general anesthesia. The individuals were randomly assigned to receive either sevoflurane or propofol throughout the induction procedure. This study's primary outcome of interest was clinical effectiveness, which was assessed by evaluating intubation ratings. Moreover, several other attributes were recorded, including patient demographics, length of laryngoscopy, ASA classification, and incidences of adverse events. **RESULTS:** The sevoflurane group had a notably greater level of clinical efficacy, as seen by 75.8% of patients obtaining successful intubation, in contrast to the propofol group, where only 51.6% of patients achieved successful intubation p-value <0.001. A significant proportion of patients in both cohorts had favorable outcomes regarding intubation ratings, ranging from satisfactory to outstanding. The incidence of adverse effects, such as laryngospasm and the administration of succinylcholine, was infrequent and seen in a minority of the patient population. CONCLUSION: Sevoflurane has been identified as a more effective induction drug for pediatric intubation without neuromuscular blockers during elective surgical procedures. The promise of sevoflurane as an alternative to conventional muscle relaxants in pediatric anesthesia is indicated by its clinical efficacy and excellent safety profile. Additional multicenter trials should be conducted to verify these results and establish sevoflurane as the preferred option for pediatric intubation.

KEYWORDS: Pediatric Anesthesia, Endotracheal Intubation, Sevoflurane, Propofol, Neuromuscular Blockers, Clinical Effectiveness, Randomized Controlled Trial.

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INTRODUCTION

The selection of induction drugs for pediatric patients is of utmost significance within the domain of contemporary anesthetic treatment ¹. Sevoflurane and propofol are two notable agents that play a significant role in the setting of

pediatric anesthesia induction ². Each of these medications has unique characteristics that contribute to the intricate process of inducing anesthesia in pediatric patients. Sevoflurane, a volatile anesthetic, has been acknowledged for its ability to promote a seamless and expeditious

induction process, along with its pleasant odor and the potential for prompt recovery ^{3.} In contrast, propofol, an intravenous anesthetic, achieves its pharmacological action by causing apnea upon injection while simultaneously reducing laryngotracheal responsiveness ⁴. This allows for the performance of laryngoscopy and intubation without the need for neuromuscular inhibition. This research's primary objective is to comprehensively compare the effectiveness of sevoflurane and propofol as intubation agents in the absence of neuromuscular blockers among pediatric patients receiving elective surgery at a tertiary care hospital ⁵.

The notion of performing endotracheal intubation in juvenile patients without producing apnea while avoiding using muscle relaxants is appealing. This technique can potentially alleviate the many negative outcomes of using muscle relaxants in the pediatric demographic. The possible difficulties discussed in this context include significant matters, including the occurrence of near-fatal bradycardia resulting from the administration of succinylcholine, as well as the complex variations seen in the recovery patterns associated with nondepolarizing drugs. It is important to acknowledge that these issues should not be disregarded, as they can substantially influence the safety and efficacy of pediatric surgical procedures ⁶.

Considerable research has been conducted to explore the methods for obtaining intubation while minimizing the associated apnea risk. Numerous research studies have extensively investigated the use of relaxant-free methods using sevoflurane, typically in combination with various supplementary agents, to facilitate the induction process and enhance the quality of intubation ⁷. The investigations have shown significant results, indicating that sevoflurane may provide benefits over propofol in laryngoscopy and intubation without the need for neuromuscular inhibition. However, the essential inquiry about the similar effectiveness of sevoflurane alone, as opposed to the widely accepted combination of propofol and succinylcholine, remains a pivotal subject of study. This aspect is especially relevant within the framework of elective surgical interventions

The importance of pediatric airway control cannot be overemphasized. The juvenile airway's distinctive anatomical and physiological features make young patients more susceptible to the rapid development of hypoxemia while experiencing apnea, hence introducing additional intricacy and possible risks to the intubation procedure. Therefore, the desire to perform endotracheal intubation in pediatric patients without causing apnea and avoiding using muscle relaxants is an attractive and possibly safer approach ⁹.

Nonetheless, the lack of scholarly investigation regarding the utilization of sevoflurane and propofol for intubation within the Pakistani community presents a significant deficiency in understanding. The variations in anthropometric characteristics across different groups need customized research studies since the results may differ. Against this crucial background, our research aimed to assess intubating circumstances using the Modified Helbo-Hansen Scoring System. The primary purpose of this study was to provide an impartial foundation for evaluating the efficacy of sevoflurane and propofol in the specific setting of pediatric patients who are scheduled to undergo elective surgical procedures.

Materials and Methods Study Design:

The present study used a randomized controlled trial methodology to evaluate the effectiveness of sevoflurane in comparison to propofol for the intubation process in pediatric patients having elective surgery, without the administration of neuromuscular blockers.

Study Setting:

The research was carried out in the Department of Anesthesiology at Patel Hospital, Karachi, for a period of six months, commencing on start date and concluding on end date.

Sampling Technique:

Non-probability consecutive sampling was used to recruit participants.

Sample Size:

The determination of the sample size was based on certain characteristics, including a confidence interval of 95%, a test power of 90%, and proportions of 96.7% for sevoflurane and 63.3% for propofol, as derived from prior research. The overall sample size was determined to be 62 children, with an equal distribution of 31 children in each group Sevoflurane and Propofol. **Sample Selection:**

Inclusion Criteria:

The research included pediatric patients with an age range extending from 1 month to 5 years. All patients in the research fell within ASA American Society of Anesthesiologists status I-II, indicating their overall physical condition and eligibility for the elective surgical operations that were scheduled under general anesthesia. Prior to the initiation of the research, informed permission was painstakingly sought from the legal guardians or parents of each child, ensuring that their participation was voluntary and that they were completely aware of the study's aims and methods.

Exclusion Criteria:

The establishment of exclusion criteria was undertaken in order to uphold the principles of research integrity and to prioritize participant safety. Participants who were older than 50 weeks post-conception, as well as those who had recently had respiratory tract infections, known medication sensitivities or allergies, or participants or guardians who declined to provide informed permission, were excluded from the study. The purpose of these criteria was to ensure the establishment of a study cohort that is uniform and medically secure, while also adhering to ethical principles in research.

Data Collection Procedure:

A controlled prospective randomized trial was conducted at Patel Hospital in Karachi, including a total of 62 patients. Following the acquisition of informed permission and the documentation of demographic information, patients were randomly allocated to either Group S Sevoflurane or Group P Propofol. The individuals received a pre-anesthetic evaluation and adhered to fasting guidelines based on their age and eating habits. Prior to induction, both groups were administered Dexamethasone and Nalbuphine intravenously. Inhalational induction with sevoflurane was administered to Group S, while Group P had intravenous induction with Propofol. The procedure of laryngoscopy and tracheal intubation was then performed, and the quality of intubation circumstances was evaluated according to pre-established criteria. The duration between the performance of laryngoscopy and the initiation of intubation was documented. In cases where intubation was unsuccessful after two tries, the administration of suxamethonium was used to facilitate a successful intubation

Data Analysis:

The data analysis was performed using SPSS version 20.0. The study computed descriptive statistics for the quantitative variables, including age, weight, and intubation score, using the mean value plus or minus the standard deviation. On the other hand, the qualitative factors, such as gender, ASA status, and efficacy, were presented as frequencies and percentages. The effectiveness of both groups was compared using chi-square tests, with a significance threshold of p < 0.05. To account for possible impact modifiers, stratification was conducted based on age, ASA status, weight, and gender. Post-stratification Chi-square tests were then used, using a significance threshold of p < 0.05. **Results**

The average age of the patients was 2.47 ± 1.64 years. The average weight of the patients was 10.85 ± 4.33 kg. The average duration from the initiation of laryngoscopy to the completion of intubation was found to be 27.98 ± 10.68 seconds. The average intubation score was 8.16 ± 3.45 Table 1.

Table 1: Mean age, weight, Time from laryngoscopy to intubation, and Intubation time of the patients

Parameters	Mean ±SD	Minimum	Maximum	
Age	2.47 ±1.64	1 month	5 years	
Weight	10.85 ±4.33	3	19	
Laryngoscopy to intubation time	27.98 ±10.68	10	53	
Intubation time	8.16 ±3.45	5	20	

This study elucidates key variables that have the potential to have a substantial influence on the anesthetic and intubation procedure. The analysis of patient distribution according to ASA American Society of Anesthesiologists status indicates that most participants were classified as ASA Status I, 61 98.4% individuals. The categorization implies that the patients exhibited

a generally satisfactory state of health, hence presenting a suitable condition for elective surgical procedures and the administration of anesthesia. Nevertheless, it is important to highlight that there was a single patient identified as ASA Status II 1, 1.6%, suggesting a greater presence of pre-existing medical conditions in this person. The consideration of laryngospasm, whether present or absent, is a significant characteristic to consider. A significant majority of patients 61 98.4% did not encounter laryngospasm, a favorable result given that laryngospasm has the potential to cause airway blockage and problems during the intubation process Table 2. An investigation was conducted on the use of succinylcholine, a pharmacological substance that induces neuromuscular blockade. A single patient 1.6% was administered succinylcholine, but most patients, 6198.6% individuals, did not get this medication Table 2. This implies that the use of succinylcholine was infrequent, maybe attributable to the effective intubation achieved without the need for supplementary muscle relaxation.

Table 2: Patient Characteristics and Clinical Outcomes

0			
Parameter	Detai	Numbe	Percentag
	1	r	e
ASA Status	Statu	61	98.4%
	s I		
	Statu	1	1.6%
	s II		
Laryngospasm	Yes	1	1.6%
	No	61	98.4%
Succinylcholin	Yes	1	1.6%
e	No	61	98.6%

The analysis of age distribution indicated that 32 51.6% individuals were aged \leq 2.5 years, while 30 48.4% were aged >2.5 years table 3. Within the cohort of patients, it was observed that 43 69.4% were males. Conversely, 19 30.6% were females Table 3. The weight distribution analysis revealed that 34 54.8% patients had a weight of 10 kg or less. Conversely, 28 45.2% participants had a weight beyond 10 kg Table 3. Table 3: Patient Characteristics by Age, Gender, and Weight

Parameters	Detail	Numbe	Percentag
		r	e
Age	< 2.5	32	51.60%
Distribution	>2.5	30	48.40%
Gender	Male	43	69.40%
	Female	19	30.60%
Weight	<10	34	54.80
Category	>10	28	45.20%
Time from	<30	43	69.40%
laryngoscopy	>30	19	30.60%
to intubation			
	One	52	83.90%
	Two	10	16.10%
Number of			
Laryngoscop			
y attempt			
Efficacy of	Clinically	47	75.80%
the patients	Effective		

carefully considering muscle relaxant dosages and intubation techniques in specific scenarios to address these conditions effectively. Furthermore, the data indicates that the distribution of Modified Helbo-Hansen scores categorized as "Excellent" 22.60% and "Good" 51.60% reflects a generally satisfactory intubation procedure. Nevertheless, the inclusion of the categories "Poor" 22.60% and "Bad" 3.20% highlights the need of doing more research to explore the variables that contribute to less desirable intubation results Table 4.

Parameters	Details	Numbe	Percentag
		r	e
Laryngoscop	Easy	35	56.0%
y Score	Fair	16	25.80%
	Difficult	9	14.50%
	Impossibl	2	3.20%
	e		
Vocal Cord	Open	28	45.20%
Score	Moving	19	30.60%
	Closing	12	19.40%
	Closed	3	4.80%
Coughing	None	40	64.50%

Table 4. I ammagagagar and Inturbation Coones	
Table 4. Larvngoscopy and initibation Scores	

Score	Slight	16	25.80%
	Moderate	5	8.10%
	Severe	1	1.6%
Jaw	Complete	46	74.20%
Relaxation	Slight	10	16.10%
Score	Stiff	4	6.50%
	Rigid	2	3.20%
Limb	None	30	48.40%
movement	Slight	16	25.80%
Score	Moderate	14	22.60%
	Severe	2	3.20%
Overall	Excellent	14	22.60%
Modified	Good	32	51.60%
Helbo-	Poor	14	22.60%
Hansen	Bad	2	3.20%
Score			

The preliminary examination of all participants reveals that Sevoflurane had superior clinical efficacy, as shown by the fact that all 31 patients 100% in this cohort achieved successful intubation. In comparison, only 16 patients 51.6% in the Propofol group were able to achieve successful intubation. The observed difference between the two groups is deemed to be statistically significant, with a p-value of less than 0.001. This finding underscores the better therapeutic efficacy of Sevoflurane. Upon further analysis, the data was divided into different age groups, leading to interesting and noteworthy discoveries. Among patients who were ≤ 2.5 years old, Sevoflurane had a success rate of 100% n=17, whereas Propofol exhibited a significantly lower success rate of 53.3% n=8. In a same manner, it was shown that Sevoflurane maintained a high level of efficacy among individuals older than 2.5 years, achieving a success rate of 100% n=14. Conversely, Propofol exhibited a success rate of 50% n=8. The observed disparities in success rates exhibited statistical significance p=0.002, indicating that the effectiveness of Sevoflurane remains stable across various age cohorts.

Additionally, a gender-focused study uncovers significant trends. In the male patient population, Sevoflurane had remarkable clinical efficacy, with a success rate of 100% n=19, which was substantially superior to Propofol 45.8%, n=11 with a p-value of less than 0.001. In contrast, while examining female patients, it was seen that Sevoflurane exhibited a notable success rate of 100% n=12, while Propofol shown a comparatively higher performance of 71.4% n=5. However, it is important to note that this difference did not reach statistical significance p=0.050 Table 5.

 Table 5: Comparison of Clinical Effectiveness
Between Sevoflurane and Propofol Groups

	Clinical ly Effectiv e	Clinicall y Ineffecti ve	Tota 1	p- value	
Comparison of effectiveness with respect to group					

Sevoflura ne	31 100	0 0	31 100	
Propofol	16 51.6	15 48.4	31 100	<0.00 1
Total	47 75.8	15 24.2	62 100	

Age ≤2.5 years and comparison of effectiveness with respect to group n=32

Sevoflura ne	17 100	0 0	17 100	
Propofol	8 53.3	7 46.7	15 100	0.002
Total	25 78.1	7 21.9	32 100	

Age >2.5 years and comparison of effectiveness with respect to group n=30

Sevoflura ne	14 100	0 0	14 100	
Propofol	8 50	8 50	16 100	0.002
Total	22 73.3	8 26.7	30 100	

Male gender and comparison of effectiveness with respect to group n=30

Sevoflura ne	19 100	0 0	19 100	
Propofol	11 45.8	13 54.2	24 100	<0.00 1
Total	30 69.8	13 30.2	43 100	

Female gender and comparison of effectiveness with respect to group n=19

Sevoflura ne	12 100	0 0	12 100	
Propofol	5 71.4	2 28.6	7 100	0.050
Total	17 89.5	2 10.5	19 100	

In the subset of patients classified as ASA Status I n=61, the clinical efficacy of Sevoflurane was shown to be exceptional, with a 100% n=31 success rate in accomplishing intubation. On the other hand, it is noteworthy that Propofol had a success rate of 53.3% n=16. The observed disparity in clinical efficacy between the two treatments is statistically significant, as shown by a p-value of less than 0.001. This finding underscores the better effectiveness of Sevoflurane specifically in patients with ASA Status I. However, the findings provide a contrasting narrative for patients classified under

ASA Status II, with a total sample size of 61. In this subgroup, neither Sevoflurane nor Propofol demonstrated successful intubation, as shown by a 0% success rate for both Sevoflurane n=0 and Propofol n=0. Although the p-value is not relevant in this scenario, the results of this study highlight the difficulties encountered when managing ASA Status II patients. It was observed that intubation posed unique obstacles irrespective of the kind of anesthetic agent used. Upon further examination of the patient cohort, it was seen that individuals falling under the weight group of 10 kg or less n=34 had a 100% n=17 success rate in intubation when administered Sevoflurane. In contrast, the success rate of intubation with Propofol in the same weight category was found to be 47.1% n=8. The observed disparity demonstrated statistical significance p<0.001, hence highlighting the efficacy of Sevoflurane within this subgroup. On the other hand, it was observed that among a group of patients n=28 with a weight more than 10 kg, Sevoflurane had a 100% success rate n=14 in maintaining its efficacy, while Propofol attained a success rate of 57.1% n=8. The observed difference between the two medicines was found to be statistically significant p=0.006. However, it is worth mentioning that both agents demonstrated rather strong performance within this subgroup Table 6

Table 6: Comparison of Clinical Effectiveness by ASA Status and Weight Category

	Clinical ly Effectiv e	Clinicall y Ineffecti ve	Tot al	p- Valu e	
ASA Status I and comparison of effectiveness with respect to group n=61					

Sevoflura ne	31 100	0 0	31 100	
Propofol	16 53.3	14 23	30 100	<0.00 1
Total	47 77	14 23	61 100	

ASA Status II and comparison of effectiveness with respect to group n=61

Sevoflura ne	0 0	1 100	1 100	
Propofol	0 0	0 0	0 0	-
Total	0 0	1 100	1 100	

Weight ≤10 kg and comparison of effectiveness with respect to group n=34

Sevoflura ne	17 100	0 0	17 100	<0.00 1

Propofol	8 47.1	9 52.9	17 100		
Total	25 73.5	9 26.5	34 100		
Weight >10 kg and comparison of effectiveness with respect to group n=28					

enectiveness with respect to group n=20					
Sevoflura ne	14 100	0 0	14 100		
Propofol	8 57.1	6 42.9	14 100	0.006	
Total	22 78.6	6 21.4	28 100		

DISCUSSION:

The administration of endotracheal intubation in pediatric patients may be achieved by using either deep inhalational anesthetic or neuromuscular blocking drugs. The mitigation of many side outcomes linked with muscle relaxants may be achieved by preserving spontaneous breathing while ensuring enough muscular relaxation for intubation. The use of this strategy has significant significance in pediatric instances, as it mitigates the occurrence of problems such as near-fatal bradycardia associated with the administration of succinylcholine in children, while also addressing the considerable variety found across individuals in terms of their recovery from nondepolarizing muscle relaxants.

The study conducted in this research showed that clinical effectiveness was reached in 47 patients, accounting for 75.8% of the total sample. Conversely, clinical ineffectiveness was detected in 15 patients, representing 24.2% of the sample. Significantly greater clinical efficacy was seen in the group receiving sevoflurane compared to the propofol group, as shown by a p-value of less than 0.001. Numerous investigations have been conducted to examine the efficacy of relaxant-free methods, including administering sevoflurane in conjunction with nitrous gas, opioids, or propofol. The primary objective of this research is to facilitate the induction process and improve the quality of intubation ¹⁰⁻¹².

The research conducted by Muhammad AK et al. 2016 determined that sevoflurane administration as a standalone agent effectively induced muscular relaxation during hernia operations in pediatric patients, therefore obviating the need for additional muscle relaxants. Furthermore, it has been shown in several studies that sevoflurane provides more favorable anesthetic conditions for laryngoscopy and intubation in the absence of neuromuscular blocking drugs, as compared to propofol ¹³⁻¹⁵.

It is noteworthy to mention that the use of sevoflurane induction without muscle relaxants, although not as efficacious as the established propofol-succinylcholine combination with an 84% rate of satisfactory intubating condition, might serve as a feasible substitute for elective operations ¹⁶.

A separate investigation examining the quality of endotracheal intubation circumstances in pediatric cleft lip and palate procedures after the administration of propofol and sevoflurane without the use of muscle relaxants revealed success rates of 96.7% for sevoflurane and 63.3% for propofol, which is consistent with the results obtained in our own research ^{17, 18.}

A research investigation carried out in the southern region of India revealed that the use of 8% sevoflurane yielded advantageous intubation circumstances during cleft operations when compared to propofol, which aligns with the findings of our study. In the same manner, research conducted in the United Kingdom revealed that the process of anesthetic induction and tracheal intubation, subsequent to a 3-minute application of sevoflurane at an 8% concentration combined with 60% nitrous oxide in oxygen, exhibited superior performance compared to intravenous induction including low-dose alfentanil and propofol ^{8, 19, 20.}

In addition, a study by Shruti et al. 2010 examined the use of sevoflurane in elective surgical procedures for pediatric patients. The findings of this investigation revealed that sevoflurane demonstrated exceptional intubation circumstances, obviating the need for neuromuscular blocking drugs ^{21.} In research done by Zhao et al. 2022 a comparison was made between sevoflurane and propofol for intubation in pediatric patients, revealing that sevoflurane had good results ²².

Furthermore, a research investigation carried out by Omara et al. 2019 examined the efficacy of sevoflurane in surgical procedures for pediatric patients with cleft lip and palate. The results of this study showed a notably high efficacy rate of 96.7% for sevoflurane, which surpassed the efficacy rate of propofol, which stood at 63.3% ²³. The findings are consistent with our own research outcomes and serve to emphasize the possible benefits associated with the use of sevoflurane in the context of pediatric intubation operations.

In research done in South India by Karanth et al. 2018, it was seen that the use of 8% sevoflurane yielded acceptable intubation circumstances during cleft procedures. This finding aligns with the outcomes of our own investigation ⁸. In research conducted in the United Kingdom, Raffy et al. 2020, discovered that the process of anesthetic induction and tracheal intubation was more effective when preceded by a 3-minute injection of sevoflurane 8% combined with 60% nitrous oxide in oxygen, as compared to intravenous induction using low-dose alfentanil and propofol ²⁴.

However, showing prudence while administering elevated dosages of intravenous propofol is essential since it may increase the propensity for producing apnea in juvenile populations. The presence of this phenomenon has the potential to undermine the benefits associated with a relaxant-free strategy, as it requires the transfer and metabolism of a greater quantity of medications from organs with a high blood supply ²⁵. Consequently, this process may delay the restoration of spontaneous breathing and the return to a conscious state. Therefore, striking a suitable equilibrium between attaining muscular relaxation and preventing excessive sedation is crucial to guarantee patient safety and achieve effective intubation ²⁶.

However, a research study examining several dosages of propofol concluded that the most effective dosage regimen consisted of a combination of 3 mg/kg of propofol administered after 3 μ g/kg of fentanyl. Moreover, recent research has proposed that administering a combination of 3 μ g/kg of fentanyl and 3 mg/kg of propofol may be a viable alternative to the established suxamethonium protocol for endotracheal intubation ²⁷.

Nevertheless, it is essential to use prudence when administering elevated dosages of intravenous propofol since it can potentially increase the likelihood of producing apnea in juvenile patients. The occurrence has the potential to undermine the benefits of a relaxantfree strategy, as it requires the transfer and metabolism of a greater quantity of medications from organs with a high blood supply. Consequently, this process may lead to a delay in the restoration of spontaneous breathing and waking. Therefore, striking a suitable equilibrium between attaining muscular relaxation and preventing excessive sedation is crucial to guarantee patient safety and achieve effective intubation.

Conclusion:

In summary, our research has shown significant findings on the effectiveness of sevoflurane compared to propofol for intubation in pediatric patients having elective procedures without using neuromuscular blockers. The findings unequivocally indicate that the use of sevoflurane in patients resulted in a notably greater degree of therapeutic efficacy. The discovery highlights the potential benefits of sevoflurane as a dependable and effective substitute for facilitating intubation in pediatric instances, especially when using neuromuscular blockers is to be circumvented.

The findings of our investigation demonstrate the clinical superiority of sevoflurane, which is consistent with the increasing body of data from previous studies. This evidence suggests that sevoflurane provides optimal intubation circumstances while reducing the need for muscle relaxants. In the field of pediatric anesthesia, it is of utmost importance to prioritize the prevention of problems related to muscle relaxants. This includes minimizing the occurrence of bradycardia and addressing the interindividual variability in medication reactions, which are highly sought-after objectives.

ETHICS APPROVAL: The ERC gave ethical review approval.

CONSENT TO PARTICIPATE: written and verbal consent was taken from subjects and next of kin.

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CONFLICT OF INTEREST: No competing interest declared.

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