

Comparison of sugammadex vs. neostigmine use in recovery of muscle relaxation related to vecuronium in obesity surgery

Obezite cerrahisinde vekuronyuma bağlı kas gevşemesinin geri dönüştürülmesinde sugammadex ile neostigmin kullanımının karşılaştırılması

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Abstract

Aim: There are many methods in anesthesia to measure the recovery after the use of neuromuscular blocking agent. Monitoring the neuromuscular function can be used to identify the residual blocks. The importance of morbid obesity in anesthesia is caused by the difficulty in succeeding sufficiency of respiration and patency of airway. There are so many methods to measure the recovery when the neuromuscular blocking agent is used. Monitoring the neuromuscular function is used to identify the residual blocks.

Methods: We designed a retrospective study with cross-sectional design. After power analysis 40 morbid obese patients operated with laparoscopic sleeve gastrectomy to treat the obesity, have been included in the study. 40 patients with sugammadex group S and neostigmine group were divided into two groups as equal.

Results: In group S, there were 14 female and 6 male having the average of 33.25 ages and the average of body mass index (BMI) as 42.96. In group N, there were 16 women and 4 men having the average of 37.55 ages and the average of BMI as 42.96. While in group S the basal train of four (TOF) value was 89.75%, in group N basal TOF was 90.65%. The duration of extubation in group S was measured 1 min and 40 sec but in group N it was measured 4 min and 39 sec. Also, the duration of recovery of muscle strength as an indicator of the recovery of basal TOF values was observed as 3 min and 15 sec in group S, and it was observed as 6 min and 18 sec in group N ($P<0.001$). The duration of extubation in group N was longer than in group S ($P<0.001$). Also, the duration of the recovery of basal TOF values as the duration of recovery of muscle strength was longer in group N than in group S ($P<0.001$).

Conclusions: While in the study with rocuronium, the duration of recovery of muscle strength with sugammadex got shorter 4 times, in our study this duration was become shorter 2 times. It can be explained as the long effects of vecuronium.

Keywords: Obesity, Sugammadex, Neostigmine, Deep neuromuscular blockade, Bariatric Surgery

Öz

Amaç: Nöromusküler bloke edici ajanın kullanımından sonra iyileşmeyi ölçmek için pek çok yöntem vardır. Nöromusküler fonksiyonun izlenmesi rezidüel blokları tanımlamak için kullanılabilir. Anestezi morbid obezitenin önemi, solunum yeterliliği ve solunum yolunun açıklığının sağlanmasındaki zorluktan kaynaklanmaktadır. Nöromusküler bloke edici ajan kullanıldığında iyileşmeyi ölçmek için pek çok yöntem vardır. Nöromusküler fonksiyonun izlenmesi rezidüel blokları tanımlamak için kullanılır. Bu çalışma veküroniyuma bağlı kas gevşemesinde sugammadexin etkinliğinin neostigmine karşı kullanımını araştırmayı amaçlamaktadır.

Yöntemler: Bu çalışmada, Pamukkale Üniversitesi Tıp Fakültesin Hastanesi'nde, obeziteyi tedavi etmek için laparoskopik sleeve gastrektomi ameliyatı yapılan 40 morbid obez hasta retrospektif olarak değerlendirildi. Hastalar, kullanılan sugammadex (Bridion®) (S grubu) ve neostigmin (N grubu) olarak iki gruba ayrıldı.

Bulgular: 40 hastanın toplanan verileri değerlendirilmesinde, S grubunda, sugammadex kullanılan 14 kadın ve 6 erkek, ortalama 33,25 yaş ve vücut kitle indeksinin (BKİ) ortalaması 42,96 olarak bulundu. N grubunda ise neostigmin kullanılan, 16 kadın ve 4 erkek mevcuttu. Hastaların yaş ortalaması 37,55 ve BKİ 42,96 idi. Hastaların ek hastalıkları yoktu. Sonuçlar değerlendirildiğinde grup S' de Bazal TOF değeri %89,75 iken, grup N'de bazal TOF %90,65 idi. Operasyon sonrası uyanma döneminde S grubundaki ekstübasyon süresi 1 dakika 40 saniye iken grup N'de 4 dakika 39 saniye ölçüldü. Ayrıca, bazal TOF değerlerinin iyileşmesinin bir göstergesi olarak kas gücünün iyileşme süresi, Grup S de 3 dakika 15 saniye grup N' de ise 6 dakika 18 saniye olarak saptandı ($P<0,001$). Grup N'deki ekstübasyon süresi, Grup S'den anlamlı olarak daha uzundu. ($P<0,001$). Ayrıca, kas kuvvetinin iyileşme süresi olarak bazal TOF değerlerinin iyileşme süresi, grup N'de grup S'den anlamlı olarak daha uzundu ($P<0,001$).

Sonuçlar: Rokuronyum ile yapılan çalışmalarda, kas kuvvetinin sugammadex ile düzelleme süresi 4 kat azalırken, çalışmamızda bu süre 2 kat azalmıştır. Bu durum ise vekuronyumun uzun etkili kas gevşetici olması olarak açıklanabilir.

Anahtar kelimeler: Obezite, Sugammadex, Neostigmin, Derin nöromusküler blokaj, Bariatrik cerrahi

Introduction

Morbid obesity (defined as body mass index >40 m²/kg) in western societies is seen 2-5% frequency and it is one of the important health problems that causes physical morbidity [1,2]. Obesity surgery is an important solution for these patients [1]. Laparoscopic sleeve gastrectomy (LSG) is a kind of surgical technique in morbid obesity treatment by the resecting of stomach fundus [3]. Recent researches showed that the success in the laparoscopic surgery is developed by deep neuromuscular blockade [4]. The importance of morbid obesity in terms of anesthesia is caused by the difficulty in sufficiency of respiration and patency of airway [2].

Neuromuscular blockers are polar and hydrophilic medicines and pharmacodynamic characters can change as a result of dispersion in adipose or lean tissue in obese patients [2]. When neuromuscular blockade is made with vecuronium in obese patients, it's observed that the duration of recovery of strength of muscle become longer. The long term effect in obese patients can be explained by relatively overdose of vecuronium in lean body mass of patient.

According to increase in dose of vecuronium, duration of neuromuscular blockade, restart of mobility and spontaneous recovery takes longer [5]. The prolonged effects of neuromuscular medicines might cause residual neuromuscular paralyzing. Potentially, length of staying in operating room and post-anesthesia care unit may be shorten [6].

There are so many methods to evaluate the recovery after use of neuromuscular blocking agent. Monitoring the neuromuscular function can be used to detect residual blocks. Neostigmine edrophonium is the most used reversal agent but the use of it is limited because of the cardiovascular and gastrointestinal side effects [7]. In 2006, in anesthesia, Gijnsberg et al. [8] published an article that is defining the first human exposure of sugammadex as modified γ -cyclodextrin developed to reverse of the blockage caused by the aminosteroid neuromuscular blocking agents, especially induced by rocuronium. After injection, sugammadex capsulizes and inactivates the rocuronium (or vecuronium) which is uncombined as tight and 1:1 dissolving complexes in water. Because of that, sugammadex creates a concentration gradient favoring the movement of rocuronium (or vecuronium) from the neuromuscular junction back into the plasma, and that results function. The fast reverse of the NMB is induced by rocuronium (or vecuronium). This mechanism of action is different from neostigmine's [9].

After releasing from nerve endings, Neostigmine can reverse the acetylcholinesterase to keep the maintenance of acetylcholine at synaptic ends, It acts as a competitive inhibitor [10,11]. As a result, acetylcholine, it pushes the neuromuscular blocking agents which are at the postsynaptic nicotinic receptors in to the competition and so neuromuscular function reappears [9]. So briefly, Sugammadex is a novel γ -cyclodextrin. It's the first a new class of selective neuromuscular blocking binding agents. Sugammadex is planned to immediately deactivate the neuromuscular blocking agents acting as acetylcholine as the most common rocuronium [7]. Sugammadex is a reliable agent for rapid recovery after mid or deep relaxation of muscle related

the use of vecuronium like use of rocuronium [6]. In this study, it's aimed to evaluate that whether sugammadex is more effective than neostigmine in recovery of muscle relaxation related to vecuronium in obesity surgery.

Materials and methods

In this study, 40 morbid obese patients operated as laparoscopic sleeve gastrectomy to treat the obesity, have been evaluated retrospectively in Pamukkale University Hospital. The patients have been separated into two groups as sugammadex (Bridion®) used (group S) and neostigmine used (group N). Before the operation, ages, gender, body mass index (BMI), basal train of four (TOF) values of patients were recorded. Patients were induced propofol 2 mg/kg after routinely given midazolam (3 mg) and premedication. Basal muscle strength values were recorded by using TOF monitoring. After 0.1 mg/kg vecuronium was applied having regard to their ideal weight and required muscle relaxation, the patients were intubated. Continuation of anesthesia was maintained by keeping inhalational anesthetic with sevoflurane and support of opioid preferred according to patient. During the operation, muscle strength parameters and additional dose of muscle relaxant requirements were determined by TOF device and additional dose was applied as needed. In obesity surgery patients, by anesthesia form, extubation duration and the duration of completely forming of postoperative muscle strength during on operation table, was recorded. The muscle strength recovery duration was calculated as minutes in which patients' basal TOF values recovered. At the end of the operation, anesthesia was ended by administering 2 mg/kg sugammadex to 20 patients, and 0.04 mg/kg neostigmine to the other 20 patients.

Statistical analysis

SPSS 15 package program was used to evaluate the data and $P < 0.05$ was accepted as statistically significant. Continuous variables were given as mean (standard deviation (SD)) (minimum value-maximum value). Chi-square test was used to compare categorical variables. T-test was used in comparison of age, BMI, basal TOF, extubation, muscle strength recovery duration parameters between groups. In order to determine the number of patients to be included in the pre-study groups, the sample size was calculated. For this study, a minimum of 36 patients was considered to be accepted in the 95% confidence interval (Graphpad StatMate 2 Windows Program). Considering the possibility of possible loss in the process due to technical reasons, a total of 40 patients were planned to be taken to the study.

Results

In group S, there were 14 female, 6 male, at the average age of 33.25 and body mass index average (BMI) of 42.96. In the N group, there were 16 female, 4 male, at the average age of 37.55 and BMI average of 42.96 (Table 1).

As the basal TOF in Group S was 89.75%, it was 90.65% in Group N. The extubation duration was detected as 1 min. and 40 sec. in group S while the duration was 4 min and 39 sec in Group N. Muscle strength recovery duration indicating the recovery of basal TOF was in 3 min 15 sec in Group S while it was 6 min 18 sec in group N ($P < 0.001$). In this study, 6 male

and 14 female in group S and 4 male and 16 male in group N, totally 40 patients were included. No significant difference was found between group S and group N in regard to age, BMI and basal TOF values ($P=0.29$). Extubation duration in Group N was significantly longer than group S ($P<0.001$). The duration of recovery of basal TOF value in group N was significantly longer than in group S ($P<0.001$) (Table 1).

Table 1: Analysis of the groups

| | Group N (n=20) | | Group S (n=20) | | P-value |
|-----------------------------------|----------------|---------|----------------|---------|---------|
| | Mean (SD) | Min-Max | Mean (SD) | Min-Max | |
| Age | 37.55 (10.10) | 19-57 | 33.25 (10.44) | 17-49 | 0.19 |
| BMI | 42.97 (4.17) | 35-51 | 42.97 (5.39) | 33-59 | 0.99 |
| Basal TOF (%) | 90.65 (3.01) | 85-96 | 89.80 (1.80) | 85-93 | 0.29 |
| Extubation Time (sec) | 279 (40.54) | 210-345 | 100.5 (27.99) | 60-165 | <0.001 |
| Muscle strength return time (sec) | 378.75 (59.58) | 285-480 | 195 (38.32) | 120-255 | <0.001 |
| Gender | n | % | n | % | |
| Female | 16 | 80 | 6 | 30 | 0.39 |
| Male | 4 | 20 | 14 | 70 | |

SD: Standard deviation, Min: Minimum, Max: Maximum, sec: Second

Discussion

The increasing prevalence of obesity in developed and developing countries and the increased medical and surgical pathologies observed in these patients necessitated the treatment of obesity [3,12]. Bariatric surgery can be applied not only to decrease in weight but also applied to reduce type 2 diabetes mellitus, hypercholesterolemia, obstructive sleep apnea (OSAS), hypertension (HT), morbidity and mortality related with obesity [3]. Nowadays, laparoscopic sleeve gastrectomy is also a frequently preferred method because of similar results in terms of weight loss and improvement of comorbidities compared to the gold standard, the gastric by-pass [3]. In the basis of laparoscopic surgery, muscle relaxation is found and deep neuromuscular blockade is required for the patients having laparoscopic sleeve gastrectomy [13]. After intraoperative deep neuromuscular blockade, the use of sugammadex shortens the leaving from the operation room and ensures the confidence of patients in the postoperative period [4]. In obesity surgery, medicine applications are performed according to the ideal weight and sometimes the medication duration takes longer when the drug is taken based on normal weight. When vecuronium is used in obese patients, the recovery duration of muscle strength increases. This prolonged duration can be attributed to many reasons in obese patients. In addition to individual variability in obese patients, prolonged elimination of vecuronium in the liver, increased neuromuscular junction sensitivity in obese patients, and over dosage according to redistribution of vecuronium in obese patients are the factors that may cause prolonged recovery [14]. If the drugs are made according to the actual weight rather than the ideal weight, a relatively overdose may occur and it can cause the muscle strength recovery be prolonged [5]. In a study, in which Weinstein et al. evaluated pharmacodynamics of vecuronium and atracurium in obese patients, it was observed that the effect of vecuronium was significantly prolonged in obese patients [14]. Each 1% increase in adipose ratio was associated with a 1.1-minute elongation in the recovery time. In addition, in a recent study, it was evaluated that if sugammadex must administer according to ideal weight or normal weight. As a result of this study, it was emphasized that sugammadex must administer as 4 mg/kg according to the ideal weight but in the

case of insufficiency of the medicine, it was emphasized that the addition of 35-50% dose must administer [13]. Also in this study, it was emphasized that this additional dose had no side effects. In our study, since the vecuronium was routinely medicated according to the ideal weight, the relative residual effect was not expected result. In our study, when we needed additional muscle relaxant, we applied additional doses according to the values determined with TOF device. Geldner et al. [6] evaluating the recovery of muscle strength for laparoscopic operations, identified that muscle relaxation caused by rocuronium and in the sugammadex group, the recovery was achieved in the first 5 minutes in 94% of patients and in neostigmine group it was found 20%. In this study, sugammadex was found 3.4 times faster than neostigmine in muscle strength recovery in normal weight patients. Laparoscopic techniques have been used in obesity surgery also in our study. It is important that performing the muscle relaxation until the end of surgery with regard to both for the comfort of the surgical area and for the safer reanimation of our patients. This is much more important in obese patients due to the difficulties airway management. In the study by Suy et al. [7] that they compared sugammadex and placebo, sugammadex was detected as effective in both patients medicated with rocuronium and vecuronium. Van Lancker et al. , in the study about whether sugammadex must medicate according to ideal or corrected weight in morbidly obese, detected that 2 mg/kg sugammadex provided a complete recovery in the ideal weight + 40% group [1]. Also Gaszynski et al. [15], in the study of comparison of sugammadex and neostigmine to detect postoperative residual curarization, they found that Sugammadex was more effective in reversing rocuronium induced muscular relaxation than neostigmine. When the duration of basal TOF in group used sugammadex was 2 min and 44 sec, it was 9 min and 37 sec in group used neostigmine [15].

In our study, when the duration of basal TOF related to vecuronium-induced muscle relaxation in obese patients used sugammadex was 3 min 15 sec, it was 6 min 18 sec in the group used neostigmine. In a study in Belgium in 2016 showed that sugammadex significantly shorten the operating room discharge and the length of intensive care unit stay [4]. Also in a multi centered study, in the USA in 2018, the use of sugammadex shortens the duration of mechanical ventilation in the intensive care unit after surgery [16]. In a recent study in Israel, it was reported that sugammadex administration improved the oxygen saturation during postoperative period [17]. Also in another study in our country in 2017, in which the ventilation functions in obesity surgery were detected, it's showed that the use of sugammadex has supportive effect on partial arterial oxygen pressure [18]. In patients undergoing laparoscopic obesity surgery, postoperative pain and analgesic therapy should be effective and should not cause adverse effects such as respiratory depression, hypoventilation and hemodynamic instability, nausea-vomiting, itching and delayed bowel function, and should allow early mobilization. A multimodal analgesic approach with different sites of action and different mechanism drugs appears to be a viable option in these cases [19]. In our study, in the case of muscle relaxation related to vecuronium in intraoperative period, the infusion of paracetamol intravenously was

administered along with intravenous administration of sugammadex or neostigmine. Sugammadex more rapidly and reliably reverses rocuronium-induced neuromuscular block compared with neostigmine but it is not known if subsequent patient outcomes, including nausea, vomiting and other aspects of recovery are modified. In the study that Peach et al. [20] studied on 304 patients, compared the recovery characteristics of sugammadex and neostigmine/glycopyrrolate following reversal of neuromuscular block. Twenty-four-hour recovery scores were not significantly different between groups. Reversal with sugammadex in this patient population did not reduce postoperative nausea or vomiting compared with neostigmine/glycopyrrolate. In the study performed with rocuronium, the muscle strength recovery duration was shortened by about 4 times with sugammadex but in our study this period was shortened by 2 times. This can be attributed to the prolonged effects of vecuronium.

We have shown that rocuronium as considered specific sugammadex can be also effective against vecuronium in the condition of absence of rocuronium in the laparoscopic sleeve gastrectomy surgery. Sugammadex is a rapid recovery agent against to blockage depending on rocuronium and vecuronium. We believe that our study will shed some light on the anesthetic management of obesity as the disease of the era.

References

1. Van Lancker P, Dillemans B, Bogaert T, Mulier JP, De Kock M, Haspelslagh M. Ideal versus corrected body weight for dosage of sugammadex in morbidly obese patients. *Anaesthesia*. 2011 Aug;66(8):721-5.
2. Arikan N, Alper I, Ulukaya S, Balcioglu ST, Yegül İ. Morbid Obez Hastalarda Rokuronyumun Farmakodinamik Etkileri. *Türk Anest Der Dergisi* 2009;37(4):234-9.
3. vanRutte PWJ, Luyer P, deHingh IHJT, Nienhuijs SW. To Sleeve or NOT to Sleeve in Bariatric Surgery? *ISRN Surgery* 2012;674042.
4. Putz L, Dransart C, Jamart J, Marotta ML, Delnooz G, Dubois PE. Operating room discharge after deep neuromuscular block reversed with sugammadex compared with shallow block reversed with neostigmine: a randomized controlled trial. *J Clin Anesth*. 2016 Dec;35:107-13. doi: 10.1016/j.jclinane.2016.07.030.
5. T. Suzuki, G. Masaki, S. Ogawa. Neostigmine-induced reversal of vecuronium in normal weight, overweight and obese female patients. *British Journal of Anaesthesia*. 2006;97(2):160-3.
6. G. Geldner M, Niskanen P, Laurila V, Mizikov M, Hu'bler G, Beck H, et al. A randomised controlled trial comparing sugammadex and neostigmine at different depths of neuromuscular blockade in patients undergoing laparoscopic surgery. *Anaesthesia*. 2012;67:991-8.
7. Suy K, Morias K, Cammu G, Hans P, van Duijnhoven WG, Heeringa M, et al. Effective reversal of moderate rocuronium- or vecuronium-induced neuromuscular block with sugammadex, a selective relaxant binding agent. *Anesthesiology*. 2007 Feb;106(2):283-8.
8. Gijsenbergh F, Ramael S, Houwing N, van Iersel T. First human exposure of Org 25969, a novel agent to reverse the action of rocuronium bromide. *Anesthesiology*. 2005;103:695-703.
9. Carron M, Zarantonello F, Lazzarotto N, Tellaroli P, Ori C. Role of sugammadex in accelerating postoperative discharge: A meta-analysis. *Journal of Clinical Anesthesia*. 2017;39:38-44. doi: 10.1016/j.jclinane.2017.03.004.
10. Naguib M. Sugammadex: another milestone in clinical neuromuscular pharmacology. *Anesth Analg*. 2007;104:575-81.
11. Mirakhur RK. Sugammadex in clinical practice. *Anaesthesia*. 2009;64(Suppl. 1):45-54.
12. Adams JP, Murphy PG. Obesity in anesthesia and intensive care. *British Journal of Anaesthesia*. 2000;85:91-108.
13. Badaoui R, Cabaret A, Alami, Y, Zogheib E, Popov I, Lorne E, et al. Reversal of neuromuscular blockade by sugammadex in laparoscopic bariatric surgery: In support of dose reduction. *Anaesth Crit Care Pain Med*. 2016 Feb;35(1):25-9. doi: 10.1016/j.accpm.2015.09.003.
14. Weinstein JA, Matteo RS, Ornstein E, Schwartz AE, Goldstoft M, Thal G. Pharmacodynamics of vecuronium and atracurium in the obese surgical patient. *Anesth Analg*. 1988 Dec;67(12):1149-53.
15. Gaszynski T, Szewczyk T, Gaszynski W. Randomized comparison of sugammadex and neostigmine for reversal of rocuronium-induced muscle relaxation in morbidly obese undergoing general anaesthesia. *Br J Anaesth*. 2012 Feb;108(2):236-9. doi: 0.1093/bja/aer330. Epub 2011 Oct 19.
16. O'Reilly-Shah VN, Lynde GC, Mitchell ML, Maffeo CL, Jabaley CS, Wolf FA. Initial experience with the unrestricted introduction of sugammadex at a large academic medical center: a retrospective observational study examining postoperative mechanical ventilation and efficiency outcomes. *Korean J Anesthesiol*. 2018 Oct;71(5):374-85. doi: 10.4097/kja.d.18.00063.
17. Evron S, Abelansky Y, Ezri T, Izakson A. Respiratory events with sugammadex vs. neostigmine following laparoscopic sleeve gastrectomy: a prospective pilot study

- assessing neuromuscular reversal strategies. *Rom J Anaesth Intensive Care*. 2017 Oct;24(2):111-4. doi: 10.21454/rjaic.7518.242.evr.
18. Piskin O, Altinsoy B, Cimencan M, Aydin BG, Okyay D, Kucukosman G, et al. The effect of bariatric anaesthesia on postoperative pulmonary functions. *J Pak Med Assoc*. 2017 Apr;67(4):561-7.
 19. Erkoç SK, Yılmaz AA. Bariatrik anestezi, postoperatif bakım ve komplikasyonlar. *Anestezi Dergisi* 2016;24:139-53.
 20. Paech MJ, Kaye R, Baber C, Nathan EA. Recovery characteristics of patients receiving either sugammadex or neostigmine and glycopyrrolate for reversal of neuromuscular block: a randomised controlled trial. *Anaesthesia*. 2018;73(3):340-7.

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