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Original Article

The effect of perineural application of bupivacaine combined with sodium bicarbonate on the synatic nerve block in rabbits after sevofluran anesthesia

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Abstract

Aim: The aim of this study was to investigate the effect of combining sodium bicarbonate with bupivacaine on prolonging peripheral nerve block time.

Material and Method: Following the approval of the required Ethics Committee, 24 male New Zealand rabbit (4250-5350 g) were randomized and divided into three groups. Group 1 sham n:8; Group 2 (bupivacaine): 0.5 mL of 0.5% bupivacaine (0.5 mg / kg) injected into the perineural area. n:8; and Group 3 (bupivacaine + sodium bicarbonate): 0.5 ml of 0.5% bupivacaine + sodium bicarbonate (125 ml of 8.4% injected into the perineural area. n: 8.

After the skin was closed in all groups, the paw pull response was monitored and recorded every 30 minutes until the sensory block of the experimental animal returned back. Hot-plate test was used for analgesia evaluation. In addition, tissue histopathology was examined for histopathological evaluation of the injection site. Sensory block was evaluated with claw tightening test and claw pull test (hot-plate) response. The measurements were carried out every 30 minutes for 120 minutes or until the block was completely resolved.

Results: 30., 60. and 90.min paw pull response in Group 2 and Group 3 showed statistically significant elongation when compared to Group 1, this difference disappeared in 120 minutes. Compared to the sham group, the 30 min hot plate and claw pull response was significantly longer in group 3 (sodium bicarbonate and bupivacaine), this difference disappeared in 60 minutes (p = 0.018).

Conclusion: When sodium bicarbonate and bupivacaine are combined, it was seen in this study that the sensory block was prolonged. We believe that the current results can be used as a guide for future studies

Keywords: Block; bupivacaine; sciatic nerve; sodium bicarbonate

INTRODUCTION

Peripheral nerve blocks are used daily as an alternative to general anesthesia and for postoperative analgesia all over the world. In addition, these blocks also reduce opioid consumption, which has serious side effects (1).

Although a single dose peripheral nerve block with long-acting local anesthetics such as bupivacain provides approximately 8-14 hours of excellent analgesia, early morning and mid-day blocks may lose their effects late at night and cause serious pain conditions. To increase the duration of peripheral nerve blocks; Drugs or adjuvants are added to long-acting local anesthetics (2,3).

Sodium bicarbonate is a drug used in alkalinization. It has been shown that the effectiveness of local anesthetics increases after alkalizing local anesthetics with sodium bicarbonate.

The aim of this study is to add more drugs and adjuvants to long-acting local anesthetics, to prolong the analgesia period more, to provide a more comfortable period by extending the pain-free period after surgery, and thus to reduce the consumption of drug with serious side effects. In our study, we investigated the effect of combining sodium bicarbonate with bupivacaine on prolonging peripheral nerve block time.

MATERIAL and METHODS

Ethics committee approval was received from Inonu University Faculty of Medicine Experimental Animals Ethics Committee for the study, dated 11.05.2017 and

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numbered 2017 / A-31. Groupings were made with 8 rabbits in each group. 24 young male New Zealand (5-6 kg) rabbits were randomized and divided into 3 groups. Group S (sham - after anesthesia with sevoflurane in the experimental animal, the sciatic nerve at the posterior extremity was revealed using a lateral incision on the calf following the separartion of the superfacial fascia. Following the exploration of sciatic nerve, 0.5ml normal saline was injected and the skin was closed without any injection of medication. n:8. Group B (0.5 mL, 0.5% bupivacain (0.5 mg / kg) was applied to the perineural area and the skin was closed.) n:8. Group BD (0.5 mL, 0.5% bupivacaine + sodium bicarbonate (20 µr / kg) was injected into the perineural area and the skin was closed.) n:8. Sevoflurane anesthesia was applied to perform the surgical procedure and post-awakening tests were started. After the skin was closed in all groups, the paw pull response was monitored every 30 minutes until the sensory block of the experimental animal returned and recorded for 120 minutes. Hot-plate test was used for analgesia evaluation. In addition, tissue histopathology was examined for histopathological evaluation of the injection site.

Histological Evaluation

The sciatic nerve segments of approximately 1-1.5 cm long were resected. For fixation, 10% neutral formaldehyde solution used for 24-48 hours. Sections were made by making horizontal and vertical axes. Tissue follow-up was applied to the tissues in a closed tissue follower, then the tissues were transformed into paraffin blocks in the tissue burial center. In the rotary microtome, 4-5 µm thick paraffin sections were prepared and deparaffinized in an oven at 60ºC. After deparaffinization, the routine hematoxylin-eosin (H&E) staining method shown below was applied to the tissues. The prepared H&E samples were evaluated under the Olympus BX-51 light microscope and photographs were taken on the Olympus DP-70 digital camera. The sham, bupivacaine and bupivacaine + dexmedetomidine groups were evaluated on the 1st and 14th days with H&E stain under the light microscope. Edema in the sciatic nerve, inflammation in the epinerium and surrounding tissues, degeneration of myelin fibers and the presence of fibrosis were evaluated semiquantitatively. Edema, inflammation, fibrosis in epinerium and surrounding tissues evaluated as follow :

0 = no edema, no inflammation and fibrosis,

1 = slight edema in small foci / mild inflammatory infiltrate / mild fibrosis,

2 = edema in local areas / moderate inflammatory infiltrate / moderate fibrosis presence,

3 = marked edema / severe inflammatory infiltrate / severe fibrosis presence)

While sciatic nerve damage evaluated as (0 = no lesion, 1 = 1-2% axon and myelin fiber damage, 2 = 2-5% axon and myelin fiber damage, 3 = fiber damage of more than 5% axon and myelin).

Statistical Analysis

Statistical analysis was performed using the SPSS program (Statistical Package for Social Sciences 21.0; SPSS, Chicago, IL, USA). Mann-Whitney U test was used for comparison between the groups. All times are given as medians (minimum - maximum). P <0.05 was considered significant.

RESULTS

Initial findings

In the hot-plate test applied for the measurement of acute thermal pain; Compared with Group S, Group B and Group BD showed significant elongation at 0, 30, 60, 90 and 120 minutes (p <0.05). When Group BD was compared with Group B, there was a significant elongation at 60 minutes (p = 0.012). There was no significant difference in other periods (Table 1).

Table 1. Hot-plate test and elongation time results, by group, for drugs[π]

Time (min)	Group S (n : 8)	Group B (n : 8)	Group BD (n : 8)	P value
Basal	11(8-16)	11(8-16)	13(7-20)	0,554
0	20(9-24)	40(10-50)α	35(35-48)β	α=0.008, β<0.001
30	11(7-23)	42(31-56)α	44(32-52)β	α<0.001, β<0.001
60	12(9-17)	39(38-58)α	54(20-55)β,*	α<0.001, β<0.001,*=0.012
90	9(5-20)	34(16-60)α	47(28-54)β	α<0.001, β=0.001
120	7(5-17)	30(28-50)α	46(20-60)β	α<0.001, β<0.001

[π]Group S: Sham, Group B: Perineural Bupivacaine (0.5 mL 0.5% (0.5 mg / kg))
Group BD: Perineural Bupivacaine (0.5 mL 0.5% (0.5 mg / kg) + Sodium Bicarbonate (20 µr / kg)
α: Significant Difference Compared to Group S β: Significant Difference Compared to Group S
* Significant difference compared to Group B

There were no significant differences between the groups in sensorial and motor block tests (Table 2).

Table 2. Motor blockade scores * for drugs, by group ¶

Time (min)	Group S (n : 8)	Group B (n : 8)	Group BD (n : 8)	P value
0	0(0-0)	2(1-2)	1(0-2)	0.667
30	0(0-0)	1(0-2)	2(0-3)	0.061
60	0(0-0)	2(1-2)	1(0-1)	0.311
90	0(0-0)	1(0-2)	1(0-3)	0.070
120	0(0-0)	1(0-1)	2(0-3)	0.352

¶ Group S: Sham, Group B: Perineural Bupivacaine (0.5 mL 0.5% (0.5 mg / kg)) Group BD: Perineural Bupivacaine (0.5 mL 0.5% (0.5 mg / kg) + sodium bicarbonate (20 µr / kg)
* Motor blockage scores (full motor blockage score = 3; normal motor function = 0)

Histological Findings

In sham group; No edema, inflammation and presence of mast cells in epineurium and surrounding tissues were observed. Inflammation in epineurium and surrounding tissue was observed on day 1 as a neutrophil dominant acute inflammatory character. Compared to the sham group in terms of edema and inflammation, there was a significant difference in Bupivacaine and Bupivacaine + dexmedetomidine group. In the Bupivacaine + dexmedetomidine group, on the 1st day compared to the Bupivacaine group, edema, inflammation in the epinerium and surrounding tissues decreased significantly. When the number of mast cells compared with the control group, there was a significant difference in the Bupivacaine and Bupivacaine + sodium bicarbonate group. In the Bupivacaine + sodium bicarbonate group, the number of mast cells increased significantly compared to the Bupivacaine group (Table 3).

Table 3. P values for histopathological evaluation, by groups, for drugs ¶

	Degeneration	oedema	Number of mast cells
1st day*	Group S & Group B	<0.01	<0.01
	Group S & Group BD	<0.01	<0.01
	Group B & Group BD	<0,05	<0.01
14th day**	Group S & Group B	<0.001	<0,05
	Group S & Group BD	<0.01	<0.001
	Group B & Group BD	>0,05	<0.05

¶ Group S: Sham, Group B: Perineural Bupivacaine (0.5 mL 0.5% (0.5 mg / kg))
Group BD: Perineural Bupivacaine (0.5 mL 0.5% (0.5 mg / kg) + sodium bicarbonate (20 µr / kg)

In the sham group, edema, inflammation and presence of mast cells in the epineurium and surrounding tissues were not observed. On the 14th day, inflammation was seen in lymphohistiocytic character. Edema in the epineurium was decreased on the 14th day compared to the Bupivacaine group in the Bupivacaine + dexmedetomidine group. In the bupivacaine + sodium bicarbonate group, inflammation in epinerium and surrounding tissues was lymphohistiocytic in character and slightly decreased. In the Bupivacaine and Bupivacaine + sodium bicarbonate group, lymphohistiocytic changes were observed in the intraneural area. when Bupivacaine group and Bupivacaine + sodium bicarbonate group compared with the sham group, edema, epinerium and increased inflammation in the surrounding tissues were statistically significant. The number of mast cells was significantly increased in the Bupivacaine + sodium bicarbonate group compared to the Bupivacaine group.

DISCUSSION

There are several studies in which rabbits are administered with sevoflurane anesthesia and sodium bicarbonate is added to bupivacaine (4-6). In the study conducted by Erdoğan et al, it was reported that, in addition to local anesthesia, perineurally administered sodium bicarbonate increased analgesic efficacy and extended waiting times in the hot-palte test, but did not cause an increase in local nerve damage and perineural inflammation (7). However, in this study, ketamine, which has high analgesic efficacy, has been used to provide anesthesia, so it has been reported that study results may be affected by this application. In addition, since awakening from ketamine anesthesia can take about 20-30 minutes, only the effectiveness after this time could be measured. However, in our study, rabbits that underwent sevaflurane anesthesia woke up within 2-3 minutes after anesthesia and measurements could be taken immediately. This is the specificity of our study. In their study, Brummet et al showed that after adding high dose sodium bicarbonate to ropivacaine, the quality of analgesia increased and the waiting time extended (8). Similarly, in another study by Brummet et al, sodium bicarbonate added to bupivacaine has been reported to cause prolongation in sensorial and motor blockade. In their study, Yektaş et al. Have shown that even if

perineurally administered sodium bicarbonate prolongs the block time, even though no local anesthetic is used in sciatic block application (9). The presence of sedative and analgesic effectiveness in intravenous administration of adjuvant drugs added to local anesthetics may affect the duration of analgesia (10-13). However, sedative and central effective analgesia efficacy of sodium bicarbonate is not expected in perineural administration, since sodium bicarbonate is administered at very low doses compared to intravenous administration and perineural absorption will be less than intravenous administration (14-18). According to the histopathological findings in the study, on the 1st day evaluation, it was found that there were significant differences in edema and inflammation in Group B and Group BD compared to Group S. However, on day 1, edema and inflammation in the surrounding tissues decreased significantly compared to Group B in Group BD. When the number of mast cells was compared, there was a significant difference in Group B and Group BD compared to Group S. Group BD was significantly higher than Group B. No mast cells were seen in Group S. On day 1, edema in Group S and inflammation and mast cell were not seen in the surrounding tissues, while edema significantly decreased in Group B compared to Group BD. In group BD, inflammation in epinerium and surrounding tissues was lymphohistiocytic and slightly decreased. The number of mast cells increased significantly in Group BD compared to Group B. No mast cells were detected in group S. Unlike our study, Erdogan et al showed that perineurally administered sodium bicarbonate did not make a significant difference in terms of local nerve damage and histopathological changes (7). This difference may have resulted from the subcutaneous saline application applied in addition to drug applications in the study by Erdoğan et al. This application may have caused more dilution in the drug (7,8). There have been no reports of sodium bicarbonate use in clinical peripheral nerve block limiting its application or with its pathological effectiveness.

CONCLUSION

As a result; In sevoflurane anesthetized rabbits,, it was observed that bupivacaine and sodium bicarbonate mixture applied to provide analgesia in sciatic block application prolongs the delay time and increases the quality of analgesia in the hot-plate test that evaluates acute thermal pain. Although sodium bicarbonate combined with local anesthetic has been shown to cause some significant changes in tissue, further histopathological evaluations are needed because it is a quantitative evaluation. The current results can be used as a guide for future studies.

Competing interests: The authors declare that they have no competing interest.

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