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Lidocaine Alkalinization with Borate Buffer: Evaluation of Onset and Duration Time of Analgesia in Peripheral Nerve Block in Rat

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Abstract

Objective: Evaluation of onset and duration of peripheral nerve blockage with alkalinized lidocaine by borate buffer instead of sodium bicarbonate.
Study design: Experimental randomized cross-over study.
Animals: Forty male Sprague-Dawley rats (body weight 200–300 g) were used in this study.
Methods: Four 1.0% lidocaine solutions were prepared from commercially available solutions. The treatment 1(n=10 rats) was lidocaine hydrochloride, treatment 2 was combination of lidocaine and borate buffer, treatment 3 (n=10 rats) was lidocaine – epinephrine, treatment 4 (n=10 rats) was lidocaine-epinephrine-borate buffer. each treatment was received 0.1 ml of the agent using a 27-G needle connected to an insulin syringe injected around the sciatic nerve. Time to the onset and duration of the superficial and deep analgesia were recorded and analyzed. The results were expressed as mean ±SD and were analyzed by a one-way analysis of variance and Duncan test as a post hoc for time of onset and duration of analgesia. Graphpad Prism version 5 software program was used for all analyses.
Results: Alkalinization of 1.0% lidocaine with or without epinephrine by borate buffer produced a faster onset than lidocaine or the combination of lidocaine and epinephrine.
Conclusion and clinical relevance: We conclude that the injection of lidocaine with added borate buffer and 1: 100,000 epinephrine increases the duration of analgesia and reduces the onset of blockade in the rat model. Further studies on the most appropriate

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dose of borate buffer are recommended. This combination might prove useful clinically to provide analgesia in infected area such as infected tooth extraction.
Key words: Lidocaine, alkalinization, borate buffer, rat

Introduction

Elevation of the pH of local anaesthetics by the addition of sodium bicarbonate was first proposed by Bignon in 1892 [1]; however, the technique has yet to find universal favour.[2] The pH of commercial local anaesthetic solutions is in the range of 4.0–6.0. Under such conditions amide local anaesthetics are largely present in an ionized state and this may influence the latency of action of these agents.[3] A number of methods exist by which the pH of local anaesthetic solutions may be increased. When the pH of an amide local anaesthetic is raised above its precipitation point there is a risk of perineural injection of insoluble particulate matter.[4-6] In addition, one study has shown previously adding sodium bicarbonate to 1% commercial lidocaine without epinephrine does not affect onset time.[7]

It was shown that alkalinization with sodium bicarbonate of 2% commercial lidocaine without epinephrine niethert affect the onset of epidural anesthesia[8], nor the onset of action of 1% lidocaine during subcutaneous injection.[9] The addition of sodium bicarbonate to 2% lidocaine produced a faster onset of epidural block [10] and of peribulbar anesthesia.[11]

The pH of a bicarbonate buffer system depends on the concentration of H_2CO_3 and HCO_3^-, and them concentration depends on the concentration of CO_2 in the gas phase, called the partial pressure of CO_2.[12] Thus in the present study the alkalinization properties of a different buffer component such as borate buffer that does not depend on the concentration of dissolved CO_2 is evaluated. Borate buffer is aqueous system that tend to resist changes in pH when small amounts of acid (H^+) or base (OH^-) are added in any metabolic condition of body.[12] The aim of this study was to evaluate onset time and duration of action of peripheral nerve blockage with new alkalinized lidocaine by borate buffer instead of sodium bicarbonate.

Materials and methods

Animals

Animal selection and management, anesthetic protocol, and preparation followed the routines approved by the Institutional Animal Care and Use Committee. Forty male Sprague-Dawley rats (body weight 200–300 g) were used in this study. They were maintained in plastic cages at the room with a 12 h-day/night cycle and an ambient temperature of 21º C, and were allowed ad libitum access to water and standard laboratory pellets. All animals used in these experiments were handled for 15 min/day for a 2-week period before the tests to preclude stress-induced analgesia in rats during experimentation.

Four 1.0% lidocaine solutions were prepared from commercially available solutions. The treatment 1 (n=10 rats) was lidocaine hydrochloride (pH=6.4., Preservative-free Lidocaine Hydrochloride, Pasteure Institute, Tehran, Iran. Treatment 2 (n=10 rats) was the combination of lidocaine and borate buffer (Sigma Chemical, St. Louis, MO., 0.15 Mol, 0.47 gr/50^{cc}), pH was adjusted to 7.76. Treatment 3 (n=10 rats) was lidocaine –epinephrine (Sigma Chemical, St. Louis, MO., 1:100,000), pH was 6.1, treatment 4 (n=10 rats) was lidocaine-epinephrine-borate buffer, the pH was adjusted to 7.65. Digital pH meter (NEL, Model 821 Turkey with Ingold Electrod U457, French) was used to determine the pH values for all solutions. There

was no sedimentation observed during the pH adjustment. The treatments injected over approximately 2 seconds with the dosage of 0.1 mL/rat.

The injection technique used in this study was the same used by Thalhammer et al. and Popitz-Bergez et al.[13,14] For injection the rat was held in lateral recumbency with the limb to be injected forming a right angle with the longitudinal axis of the trunk. The greater trochanter and ischial tuberosity were localized by palpation. On an imaginary line from the greater trochanter to the ischial tuberosity, about one third of the distance caudal to the greater trochanter, the injection needle was advanced from dorsolateral direction at a 45 degrees angle until the tip encountered the ischium. Then 0.1 ml of each treatment injected using a 27-G needle connected to an insulin syringe. Analgesia was measured in the limb every 2 minutes after the injection for up to 40 minutes, and every 10 minute thereafter. Nociception was quantified by evaluating the rat's withdrawal response to apply pressure across a skin fold over the lateral metatarsus (superficial cutaneous pain) or across the distal phalanx of the first and fifth toe (deep pain) by a force calibrated forceps with a tip diameter of 2 mm [13]. The withdrawal response was graded on an ordinal scale of 0 (no withdrawal response) to 4 (a normal, brisk withdrawal response). A score of 4 meant a normal reaction characterized by a brisk, strong paw withdrawal, vocalization, and an attempt to bite the forceps. A score of 3 was characterized by a slower, weaker withdrawal response, vocalization, and no attempt to bite the forceps. A score of 2 corresponded to an even slower withdrawal response, no vocalization, and no biting of the forceps. A score of 1 was characterized by a very weak attempt to withdraw. And a score of 0 was given when the rat showed none of these responses. Previous reports showed that motor block of the sciatic nerve could not account for withdrawal response deficits, proving that true sensory loss was being tested [13]. Time to onset and duration of the superficial and deep analgesia were recorded. Time from the injection to loss of the sensation (up to score 0) was considered as time of the onset of the analgesia. Time between loss (score 0) and reappearance of pain response (score 4) was considered as duration time of analgesia.

The results were expressed as mean ±SD and were analyzed by a one-way analysis of variance and Duncan test as a post hoc for time of onset and duration of analgesia. Graphpad Prism version 5 software program was used for all analyses. A value of $P < 0.05$ was considered significant.

Results

Considering onset time of superficial and deep analgesia, alkalinization of 1.0% lidocaine with or without epinephrine by borate buffer produced a faster onset than unalkalinized lidocaine or the combination of lidocaine and epinephrine (Table 1). Furthermore, onset times with either alkalinizing agent did not differ significantly from each other (P= 0.3, Table 1).

Considering duration of superficial and deep block, these parameters did differ significantly among treatments 1 and 2 with treatments 3 and 4. Longer duration of analgesia was recorded for treatment 3 and 4 in comparison with treatment 1 and 2 (P<0.05, Table 1). There were no significant differences between treatment 3 and treatment 4 in duration of analgesia (P>0.05, Table 1).

Table1. Anesthetic indices of sciatic nerve block with four treatments in rat (mean ± SD)

Treatment type	Superficial analgesi		Deep analgesia	
	Onset time (second)	Duration time(minute)	Onset time (second)	Duration time (minute)
Treatment 1 (lidocaine)	138.0±16.4[a]	45.2±4.5	210.0±36.7[c]	42.0±4.2
Treatment 2 (lidocaine buffer)	60.0±21.2	52.2±12.8	108.0±16.0	49.2±11.7
Treatment 3 (lidocaine –epinephrine)	153.7±43.7[a]	79.1±18.6[b]	270.0±60.0[c]	75.1±19.1[d]
Treatment 4 (lidocaine –epinephrine-buffer)	67.5±26.6	72.7±11.8[b]	138.7±42.2	70.7±11.6[d]

[a] statistical difference ($p<0.05$), between onset time of superficial analgesia in treatment 1 and 3 with treatment 2and 4. Onset time in treatment 2 and 4 were faster than treatment 1 and 3.
[b]statistical difference ($p<0.05$) between duration time of superficial analgesia in treatment 3 and 4with treatment 1 and 2. Duration of analgesia were longer in treatment 3 and 4.
[c]statistical difference ($p<0.05$), between onset time of deep analgesia in treatment 1 and 3 with treatment 2and 4. Onset time in treatment 2 and 4 were faster than treatment 1 and 3.
[d]statistical difference ($p<0.05$) between duration time of deep analgesia in treatment 3 and 4 with treatment 1 and 2. Duration of analgesia were longer in treatment 3 and 4.

Discussion

The degree of nerve blockade was enhanced by alkalinized lidocaine. An increase in the nonionized fraction of the local anesthetic (which is associated with an increase in pH) results in improved nerve penetration and a more rapid onset of nerve blockade [15,16], this phenomena was shown in our study that alkalinized lidocaine with borate buffer with or without epinephrine has more significant fasten onset of time of superficial and deep analgesia in comparison of unalkalinized forms.

It was shown that alkalinization with sodium bicarbonate of 2% commercial lidocaine without epinephrine neither affect the onset of epidural anesthesia [8], nor the onset of action of 1% lidocaine during subcutaneous injection.[9] However, other studies have reported conflicting results. It was shown that alkalinization with sodium bicarbonate does accelerate the onset time of lidocaine. The addition of sodium bicarbonate to 2% lidocaine produced a faster onset of epidural block in one study [10] and of peribulbar anesthesia in another.[11] None of the clinical studies include pH of the injected soloutions, before or after adjuvant addition. This is to point out that borate buffer is not approved for clinical use as an alkalinizing agent for local anesthetics and do not advocate its usage for this purpose until rigorous clinical testing of its safety and efficacy is performed. The results of this study indicated that alkalinized lidocaine with borate buffer did not decrease the block duration of lidocaine without epinephrine in comparison with plain lidocaine. This is in agreement with the results of Parham et al. study.[10] They showed that adding sodium bicarbonate to 1% lidocaine without epinephrine did not affect its duration of action after subcutaneous injection. Precipitation was considered as a possible explanation for the effect of sodium bicarbonate on lidocaine's duration of action.[5] However, no precipitate was observed by visual inspection during any component of our study. Furthermore, lidocaine adjusted to the identical pH with borate buffer did not decrease the duration of analgesia in comparison with plain lidocaine, indicating that precipitation probably did not occur. Because lidocaine solutions at the same pH have the same concentration of the base form of the local anesthetic and it is expected to precipitate at the same rate at the same pH. In the present study adding borate buffer to commercial 1% lidocaine with epinephrine (1:100,000) did accelerate the onset. This may

result from elevation of the low pH (6.1 to 7.65) of the 1% solution, a situation that often occurs with epinephrine- containing solutions and that would be expected to accelerate block onset.
It is concluded that the injection of lidocaine with added borate buffer and 1: 100,000 epinephrine increases the duration of analgesia and reduces the onset of blockade in the rat model. Further studies on the most appropriate dose of borate buffer are recommended.

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چکیده

لیدوکائین قلیایی شده با بافر بورات: ارزیابی شروع و طول اثر بیدردی در بی حسی عصب محیطی در موش صحرایی

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هدف- ارزیابی شروع وطول اثر بیحسی عصب محیطی با لیدوکائین قلیایی شده با بافر بورات بجای بی کربنات سدیم
طرح- مطالعه تجربی
حیوانات- ۴۰ قطعه موش صحرایی نر بالغ به وزن ۳۰۰-۲۰۰ گرم در این مطالعه استفاده شدند
روش- چهار ترکیب ۱٪ از لیدوکائین تجاری موجود آماده سازی شد. در گروه درمانی ۱ (به تعداد ۱۰ قطعه) لیدوکائین هیدروکلراید، در گروه درمانی ۲ (به تعداد ۱۰ قطعه)، لیدوکائین- بافر بورات، در گروه درمانی ۳ (به تعداد ۱۰ قطعه) لیدوکائین- اپی نفرین و نهایتا در گروه درمانی ۴ (به تعداد ۱۰ قطعه) لیدوکائین- اپی نفرین-بافر بورات استفاده شد. ۰/۱ میلی لیتر از هر ترکیب با سر سوزن گاج ۲۷ با سرنگ انسولین در اطراف عصب سیاتیک تزریق شد. زمان شروع و طول بیدردی سطحی و عمقی ثبت شده و مورد آنالیز آماری قرار گرفت
نتایج- شروع اثر بیدردی سطحی و عمقی با لیدوکائین- بافر بورات با اپی نفرین یا بدون اپی نفرین سریعتر از لیدوکائین قلیایی نشده و یا لیدوکائین- اپی نفرین بود. اضافه کردن اپی نفرین در گرو ههای مربوطه باعث افزایش طول اثر شد.
نتیجه گیری- از این مطالعه چنین نتیجه ای می توان گرفت که قلیایی کردن لیدوکائین با بافر بورات و اضافه کردن ۱ به ۱۰۰۰۰۰ اپی نفرین باعث شروع اثر سریع و طول اثر بیشتر در مدل حیوانی موش صحرایی می شود. مطالعات بعدی لازم است که در مورد دوزهای مختلف بافر بورات صورت گیرد. احتمالا از این ترکیب می توان در محیط های اسیدی مثل محیط های عفونی مثل آبسه های دندانی استفاده نمود که نیاز به مطالعه در این زمینه می باشد که ما در این مطالعه در این مورد هیچ گونه ارزیابی انجام نداده ایم.
کلید واژگان- لیدوکائین، قلیایی کردن، بافر بورات، موش صحرایی.