Title: Magnesium sulfate attenuates tourniquet pain in healthy volunteers

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Abstract:

Purpose: Preoperative administration of *N*-methyl-D-aspartate (NMDA) receptor antagonist has been shown to attenuate tourniquet-induced blood pressure increase under general anesthesia, suggesting that the mechanism includes NMDA receptor activation. This attenuation may be associated with pain relief caused by NMDA receptor antagonism. We tested the hypothesis that magnesium sulfate, an NMDA receptor antagonist, attenuates tourniquet pain.

Methods: Twenty-four healthy volunteers were randomly assigned to four groups (n=6 each): control (normal saline), M1 (magnesium, 1 g), M2 (magnesium, 2 g) and M4 (magnesium, 4 g). Normal saline or magnesium solution was given intravenously over 15 min in a double-blind fashion before tourniquet inflation, which was continued for 60 min or until ”pain score” (0 = no pain, 100 = highest tolerable pain) reached 100. Pain scores were recorded before and every 5 min during inflation. If subjects reported a pain score of 100 before the end of the 60-min period, we adopted a pain score of 100 for the remaining period.

Results: Duration of inflation in the M4 group was significantly longer than in the control group (54.3 ± 8.3 *vs.* 42.9 ± 9.9 min, *P*=0.03). Pain scores in the M4 group were significantly lower than in the control group from 10 through 50 min after the start of tourniquet inflation. Area under the curve for pain scores in the M4 group was significantly

smaller than in the other groups.

Conclusion: Magnesium sulfate, 4 g, significantly attenuated tourniquet pain in healthy awake volunteers, suggesting that NMDA receptor activation is involved in tourniquet pain.

Introduction

Tourniquets are widely used during orthopedic surgery involving the upper or lower limbs to minimize bleeding and keep the surgical field dry. However, this custom is sometimes accompanied by so-called tourniquet pain and tourniquet-induced arterial blood pressure (BP) increase. Tourniquet pain is characterized by a gradual onset of dull aching at the site of the tourniquet or distal extremity despite otherwise adequate regional anesthesia, occasionally necessitating supplemental general anesthesia [1]. Tourniquet-induced BP increase may develop after the start of tourniquet inflation and these changes follow the same time course as tourniquet pain [1,2].

Ketamine, dextromethorphan, and magnesium are antagonists of the *N*-methyl-D-aspartate (NMDA) receptor [3]. We reported that low-dose intravenous ketamine attenuates tourniquet-induced BP increase under general anesthesia [4]. Dextromethorphan [5] and magnesium [6] have also been reported to have this effect. These results suggest that the mechanism of tourniquet-induced BP increase includes NMDA receptor activation.

Attenuation of tourniquet-induced BP increase may be associated with pain relief due to NMDA receptor antagonism. However, few data are available that directly link tourniquet pain to NMDA receptor activation. We therefore tested the hypothesis that magnesium sulfate, which, in contrast with ketamine [7], does not have a direct analgesic effect, would attenuate tourniquet pain in healthy awake volunteers.

Materials and methods

The study protocol was approved by our ethics committee and performed in a randomized, double-blind, prospective fashion. We explained that volunteers could ask to deflate tourniquet when they felt the pain intolerable. Written informed consent was obtained from each volunteer. Twenty-four healthy male volunteers, ASA physical status class I, aged 22 to 23 years, were recruited to the study. Exclusion criteria were hypertension, ischemic heart disease, deep vein thrombosis, and a past history of chronic pain, mental disorder, or substance misuse. Subjects were randomly assigned using computerized random number generation, to one of four groups (described below) with six volunteers in each. A staff member not involved in the study prepared identical bags containing each test solution. One of the authors, who was blinded to the test solution, collected data from all volunteers.

During the experiment, BP (measured noninvasively), heart rate (HR), electrocardiography, and oxygen saturation were monitored. The following test solutions were used in each group, to give a total volume of 100 ml in each: control group (n=6), normal saline; M1 group (n=6), 10 ml of 10% magnesium sulfate (1 g) diluted with saline; M2 group (n=6), 20 ml of 10% magnesium sulfate (2 g) diluted with saline; and M4 group (n=6), 40 ml of 10% magnesium sulfate (4 g) diluted with saline. Saline and magnesium solutions were similar in appearance. After securing venous access from the dorsal venous network of the hand or the cephalic vein, the test solution was given over 15 min before tourniquet inflation. Pain intensity was evaluated using a 100 mm visual analogue scale

(“pain score”: 0 = no pain, 100 = highest tolerable pain). An 11-cm wide standard orthopedic tourniquet was inflated at the thigh level to 400 mmHg, and this was continued for 60 min or until volunteers expressed that they could not tolerate the pain any more (“pain score” = 100). Venous blood was sampled to measure serum magnesium concentration before and after the experiment.

Pain score, BP, and HR were recorded every 5 min during tourniquet inflation. If the subjects reported a pain score of 100 before the end of the 60-min period, we adopted a pain score of 100 for the remaining time. We also recorded the duration of tourniquet inflation (tourniquet time). Side effects attributed to magnesium toxicity (nausea, sedation, cardiac conduction disorder, and hypoventilation) were also recorded during the study period. Volunteers were instructed to report any symptoms, such as nausea, drowsiness, muscle weakness, or dyspnea. Furthermore, at the end of experiment a blinded observer asked volunteers whether they had experienced these symptoms. During the experiment, cardiac conductive function was assessed using ECG and respiration was monitored using pulse oximetry by the blinded observer.

Demographic data, tourniquet time, and serum magnesium concentration were expressed as mean ± SD and compared among the four groups using one-way analysis of variance (ANOVA) with Bonferroni’s correction. Hemodynamic data were analyzed by repeated-measures ANOVA. In addition, BP and HR were statistically compared with their baseline values using Student’s t-test. Changes in pain scores were compared among the

four groups using repeated-measures ANOVA. When statistical significance was found, *post-hoc* comparisons were made by Bonferroni's method. The area under the curve (AUC) for pain scores was determined and compared using one-way ANOVA with Bonferroni's correction. *P* values of less than 0.05 were considered statistically significant. Sample size was determined assuming that AUC for pain scores was 2400 in the M4 group and 4000 in the control group. The required numbers for each group in this study were calculated using power analysis to find a significant difference of $P<0.05$ ($\alpha=0.05$) with a power of 90% (β error=0.1). This analysis determined 6 volunteers per group as sufficient. The primary outcome measure of this study was the effect of magnesium sulfate in attenuating tourniquet pain.

Results

The four groups were similar in terms of age, height, and weight (Table 1). Tourniquet time in the M4 group (54.3 ± 8.3 min) was significantly longer than in the control group (42.9 ± 9.9 min) ($P=0.03$) (Table 1). Serum magnesium concentrations before the experiment were similar among the four groups. At the end of the study, volunteers in the M1, M2 and M4 groups had significantly increased serum magnesium concentrations compared with before the experiment (Table 1). There was no significant difference in baseline blood pressure between groups. No significant hemodynamic differences among the four groups were observed over time. However, there was a significant systolic BP increase 30 min after

tourniquet inflation in the control (P=0.03), M1 (P=0.03), and M2 (P=0.01) groups, which was not seen in the M4 group (P=0.22) (Table 2).

Pain scores showed significantly less change over time in the M4 group than in the other three groups (Figure 1). Pain scores in the M4 group were significantly smaller than in the control group from 10 to 50 min after tourniquet inflation (Figure 1). In the M4 group, AUC for pain scores (2400 ± 939) was also significantly smaller than in the other three groups (control: 3952 ± 291, M1: 3438 ± 440, M2: 3740 ± 745).

During the study period, no electrocardiography change was observed and oxygen saturation was kept more than 97%. No symptoms of magnesium toxicity were reported.

Discussion

We found that magnesium sulfate, at a dose of 4 g, significantly delayed reaching a pain score of 100, attenuated tourniquet pain, and prevented systemic BP increase during tourniquet inflation in healthy awake volunteers.

The etiological factors and neural pathways involved in tourniquet pain remain controversial [8]. In this study, magnesium attenuated tourniquet pain despite not having a direct analgesic action. Magnesium is a physiological antagonist of calcium channels [9] and NMDA receptors [10], and these actions are considered important in its antinociceptive effect [11,12]. Tourniquet inflation activates C fibers [13,14], and prolonged firing of C fiber nociceptors activates NMDA receptors in the spinal cord, initiating a series of central

sensitization changes such as the wind-up phenomenon and long-term potentiation [15]. Such central neuronal plasticity has an important role in pathological pain perception, and its induction and maintenance depends on the NMDA receptor [12]. It is possible that tourniquet pain is caused by acute-onset temporary hyperesthesia via central neuronal plasticity. Voltage-dependent and noncompetitive block of the NMDA receptor by magnesium ions probably attenuates persistent pain by inhibiting calcium influx into the cell [10] and suppressing central sensitization resulting from persistent inputs of noxious stimuli. However, further confirmation of this mechanism is required. In fact, magnesium is reported to reduce neuropathic pain probably by blocking NMDA receptor [16].

Other mechanisms are possible. Intra-articular magnesium has been found effective for postoperative analgesia in arthroscopic knee surgery [17,18], suggesting that antagonism of peripheral NMDA receptors prevented peripheral sensitization [19,20]. This action might accordingly be associated with the reduction of tourniquet pain in the present study. Moreover, magnesium protects the central nervous system from ischemic damage [21], such as that caused by tourniquet inflation.

The effective magnesium concentration in the serum and cerebrospinal fluid (CSF) necessary for antagonism of central NMDA receptors remains unclear. Magnesium suppresses neuropathic pain responses via a spinal site of action in rats [22]. Although intravenous magnesium administration leads to a significant and long-term increase in CSF magnesium concentration, the magnitude of this increase is small [23], indicating that a

high dose of systemic magnesium is needed for spinal analgesia. A high dose of magnesium, 4 g, was also necessary to reduce tourniquet pain in our study. We accordingly agree with Hwang et al. that adequate doses of magnesium sulfate are important for effective analgesia [24].

In general, symptoms of magnesium toxicity develop at serum magnesium concentrations above 2 mmol/L [24]. In this study, the serum magnesium concentration after the experiment in the M4 group (1.54 ± 0.10 mmol/L) was below this level, and no symptoms of magnesium toxicity were observed. A higher dose of magnesium sulfate is used for the treatment of pre-eclampsia [21] and did not produce sedation or reduce muscle strength in healthy volunteers [25]. Thus, 4 g of magnesium sulfate seems to be a reasonable dosage; however, special regard should be paid when administering this drug to patients with renal dysfunction.

This study has some limitations. Firstly, although NMDA receptor activation is suggested to be involved in the mechanism of tourniquet pain, we could not obtain conclusive evidence of this. Further basic animal experiments are needed to clarify the more precise mechanism of tourniquet pain. Secondly, the dose of magnesium was not adequate to prevent tourniquet pain. The maximum magnesium dose was set at 4 g for ethical reasons. While larger doses of magnesium sulfate might be more effective in attenuating tourniquet pain, the risk of side effects or toxic symptoms would be heightened. Thirdly, we recruited only male participants because the menstrual cycle influences the

experience of pain [26]; we believed that limiting the study to men would allow us to gather data under more uniform conditions and evaluate the effect of magnesium more precisely. Finally, close age and body weight of volunteers and the risk of magnesium toxicity prevented us from determining the optimal dose of magnesium.

In conclusion, magnesium sulfate, at a dose of 4 g, significantly reduced tourniquet pain. Our findings suggest that NMDA receptor activation is involved in the mechanism of tourniquet pain.

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Table

Table 1. Volunteers' demographic data, the duration of tourniquet inflation, and serum magnesium concentrations.

Group	Control group	M1 group	M2 group	M4 group
Number of volunteers	6	6	6	6
Age (years)	22.2 ± 0.4	22.2 ± 0.4	22.3 ± 0.5	22.5 ± 0.5
Height (cm)	171.0 ±2.8	171.0 ±4.6	174.3 ± 2.8	172.5 ± 5.5
Weight (kg)	66.0 ± 6.0	65.0 ± 4.1	66.7 ± 3.9	62.3 ± 5.5
Tourniquet time (min)	42.9 ± 9.9	44.7 ± 4.3	42.5 ± 10.1.	54.3 ±8.3*
Mg concentration (mg/dl): PRE	2.3 ± 0.1	2.3 ± 0.1	2.3 ± 0.1	2.3 ± 0.1
Mg concentration (mg/dl): POST	2.2 ± 0.1	2.8 ± 0.3*	3.3 ± 0.2*	3.8 ± 0.3*

Values are presented as mean ± SD.

Tourniquet time, the duration of tourniquet inflation; PRE, before the experiment; POST, after the experiment

*P<0.05 compared with the Control group.

Table 2. Volunteers' hemodynamic data.

Variable	Group	Baseline	30 min	After deflation
Systolic AP (mm Hg)	Control	128.2 ± 13.7	139.5 ± 14.7*	132.8 ± 19.3
	M1	117.0 ± 8.1	130.0 ± 9.6*	129.8 ± 6.2*
	M2	125.2 ± 15.0	139.7 ± 16.1*	140.2 ± 14.7
	M4	115.7 ± 6.5	125.5 ± 13.9	118.0 ± 13.7
Diastolic AP (mm Hg)	Control	71.8 ± 8.9	85.5 ±14.1*	73.5 ± 8.5
	M1	74.0 ± 8.2	87.5 ± 6.5*	79.2 ± 9.7
	M2	69.5 ± 7.5	85.3 ±6.0*	73.2 ± 18.7
	M4	62.8 ± 5.6	77.3 ± 6.6*	57.3 ± 15.9
HR (bpm)	Control	73.7 ± 14.2	76.3 ± 12.6	72.8 ± 14.8
	M1	70.7 ± 7.4	69.5 ± 7.0	76.3 ± 9.0
	M2	67.7 ± 3.9	71.0 ± 7.3	72.8 ± 7.7
	M4	71.7 ± 9.6	74.7 ± 12.4	69.3 ± 14.7

Values are expressed as mean ± SD.

30 min = 30 minutes after the start of tourniquet inflation

AP = arterial blood pressure; HR = heart rate.

*P<0.05 compared with Baseline.

Figure Legends

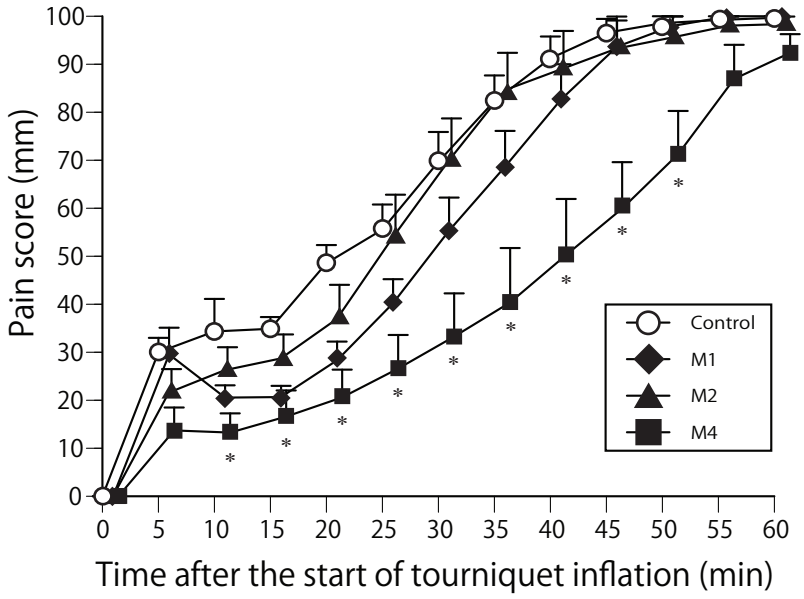
Figure 1:

Intensity of pain after the start of tourniquet inflation using a 100 mm visual analogue scale (Pain score: 0 = no pain, 100 = highest tolerable pain).

Pain scores in the M4 group were significantly lower than in the control group (*$P < 0.05$ *vs.* control group).

Data are presented as mean and error bars represent the standard deviations.

Fig. 1


Pain score (mm)
100
90
80
70
60
50
40
30
20
10
0
0
5
10
15
20
25
30
35
40
45
50
55
60
Time after the start of tourniquet inflation (min)
Control
M1
M2
M4
*