Sleep Deprivation Reduces Tear Secretion and Impairs the Tear Film

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Purpose. We investigated whether sleep deprivation (SD) disturbs the tear film.

Methods. A total of 20 healthy male subjects with no ocular disease was recruited: 10 were allocated to the SD group and 10 to the control group The 10 subjects in the SD group were deprived of sleep in an experimental setting and their outcomes were compared to those of the control group, which was not sleep-deprived. Tear film and ocular surface were evaluated at 2 PM, 10 PM, and 6 AM and 2 PM the following day. Tear osmolarity, Schirmer's test, tear film break-up time (TBUT), pain on a visual analog scale (VAS), and IOP were measured.

Results. At 6 AM the following day, mean tear osmolarity level increased ($P = 0.004$), TBUT was significantly shorter ($P = 0.01$), and tear secretion measured by Schirmer's test was significantly reduced in the SD group than in the control group ($P = 0.004$). No significant change in IOP was observed in either group.

Conclusions. Sleep deprivation induced tear hyperosmolarity, shortened TBUT, and reduced tear secretion, all of which can trigger the development of ocular surface diseases. Therefore, SD can exacerbate signs and symptoms in patients with ocular surface diseases. (ClinicalTrials.gov number, NCT02026986.)

Keywords: sleep deprivation, tear hyperosmolarity, tear break-up time, tear secretion

Tear film consists of three layers, including the outer lipid layer, aqueous layer, and inner mucin layer.[1,2] The lipid layer protects the aqueous layer of tear film from evaporation, while the mucin layer adheres the tear film to the ocular surface. The aqueous layer, which is produced in lacrimal glands, is the most important layer with respect to ocular surface health. Reduction of aqueous tear secretion or increased evaporation of tear film induced by disruption of the lipid layer results in tear film instability and in the disruption of homeostasis at the ocular surface, and leads to dry eye syndrome.[2] Dry eye syndrome is a common ocular surface disease associated with symptoms of eye discomfort, grittiness, and visual disturbance.[1,2] Dry eye syndrome disrupts normal homeostasis at the ocular surface, resulting in epithelial damage, epithelial cell apoptosis, loss of goblet cells, and squamous metaplasia.[1–3] The changes and inflammation of the ocular surface subsequently lead to tear instability, which causes increased tear osmolarity and aggravates the inflammatory cascades. This leads to a vicious cycle.[2] The regulation of tear film secretion is under neural and hormonal control.[4] Dry eye syndrome has been associated with diverse and multiple causes, including depressive disorders, drug uses, hormonal status, and systemic diseases.[2]

Sleep deprivation (SD) is known to cause profound impairments in executive function and vigilant attention.[5,6] It also reportedly is associated with autonomic and endocrine functioning,[7–9] and has been shown to increase blood pressure and stress hormone levels, and decrease parasympathetic tone.[10,11] Tear secretion is regulated by neurologic factors and hormones,[12] and, thus, SD may have an effect on the tear film and ocular surface. However, only a few studies have evaluated the effect of SD on tear film and the ocular surface. We investigated this effect in this study,

Methods

We enrolled in this study 20 healthy young male volunteers aged 20 to 30 years. The decisive criterion for inclusion in the study was a normal body mass index (BMI) of 20 to 25. All subjects were required to complete sleep questionnaires. Only subjects with a self-reported sleep length of 7 to 7.5 hours were enrolled. The subjects were interviewed to determine their medical history and underwent a physical examination before the experiment. The medical history interview and physical examination revealed no signs of systemic disease, history of ophthalmic surgery, or dry eye symptoms within the preceding 6 months. The subjects were not on any medication and none was using eye drops. Subjects with dry eye symptoms within the previous 6 months were excluded from the study. Subjects who had any systemic diseases, such as systemic lupus, rheumatoid arthritis, Sjögren's syndrome, or a history of ocular disease, and disorders of the lid margin, nasolacrimal duct, and cornea were excluded. All of the subjects were good sleepers and nonhabitual nappers with no excessive daytime sleepiness. The subjects were divided randomly into two groups: the SD

and control groups. Ten subjects in the SD group were examined after a SD experiment in which they did not sleep for 24 hours. The 10 subjects in the control group were not sleep deprived (they had 8 hours of sleep) and were examined using the same protocol, which was approved by the Institutional Review Board of Hallym University Medical Center and adhered to the Declaration of Helsinki. All subjects signed an informed consent form before participating in the study.

Testing Protocols

The experiments began at 2 PM, and the subjects were examined at four time points: 2 PM and 10 PM, and at 6 AM and 2 PM the following day. All subjects were evaluated by the same investigator (YBL) within 1 hour. Tear osmolarity, subjective symptoms assessed by visual analog scale (VAS), tear film break-up time (TBUT), and performance on Schirmer's tear secretion test were evaluated. Subjects in the SD group did not sleep for one entire day and subjects in the control group went to bed at 10 PM. The sleep time in the control group was set from 10 PM to 6 AM the following day. None of the subjects was allowed to wash or apply eye drops. In addition, they did not participate in any activities that could influence the ocular surface and tear film, such as watching TV or reading a book. Water intake was not restricted during sleep deprivation. Running or sports activities that induce excessive sweating or stimulating the autonomic nerve system were restricted. The investigator strictly observed all subjects throughout the study period. All subjects underwent the same examinations, including measurement of tear osmolarity, assessment of subjective symptoms using a VAS, Schirmer's tear test, TBUT, and ocular surface fluorescence staining at 2 PM and 10 PM, and at 6 AM and 2 PM the following day.

Tear Osmolarity Measurement

Tears were collected as described previously.[10] Briefly, a microcapillary glass tube (Marienfeld, Lauda-Königshofen, Germany) was placed on the lower outer conjunctival sac. To avoid reflex tearing, the subjects were asked to direct their gaze supranasally. A total of 30 μL of tears was obtained from the marginal tear strip. After centrifugation at 1000 *g* for 3 minutes, supernatants were obtained, and the samples were stored at −80°C. Tear osmolarity was measured using a Multi-OSMETTE 2430 (Precision Systems, Inc., Natick, MA, USA).

TBUT Evaluation

The TBUT evaluation was performed in a dimly lit room. Fluorescein was placed in the lower conjunctival sac using a fluorescein strip (Haag-Streit, Köniz, Switzerland). The subjects were asked to blink, and the time before the first defect appeared on the stained tear film was recorded as the TBUT.[3,10]

Schirmer's Tear Secretion Test

Schirmer's test was performed to evaluate the effect of SD on tear secretion. Filter papers (Color Bar; EagleVision, Memphis, TN, USA) were placed in the lateral canthus for 5 minutes; readings were recorded as millimeters of wetting.[3,10]

Visual Analog Pain Scale

Subjective symptoms were graded numerically using a VAS.[10] The scale ranged from 0 (absence of pain) to 10 (maximum degree of pain). The subjects were asked to describe their discomfort or pain using the VAS at each preset time.

IOP Measurement

The IOP was measured by noncontact tonometer (CT-80; Topcon Corp., Tokyo, Japan). Intraocular pressure was expressed in millimeters of mercury (mm Hg).

Statistical Analysis

Analyses were performed with SPSS software, version 12.0K (SPSS, Inc., Chicago, IL, USA). The Wilcoxon signed rank test and Mann-Whitney *U* test were used to compare the tear film and ocular surface changes. A P value of <0.05 was considered statistically significant.

Results

The characteristics of the subjects at baseline are summarized in Table 1. The mean age of control group and SD group was 24.57 ± 1.31 years and 25.25 ± 1.04 years. There was no significant difference between two groups in VAS, TBUT, Schirmer's test, tear osmolarity, and IOP.

Tear Osmolarity Measurement

Tear osmolarity in the SD group increased from baseline to 6 AM the following day ($P = 0.008$, Wilcoxon signed rank test), but returned to baseline by 2 PM that day. Tear osmolarity at 6 AM the following day was lower in the SD group compared to the control group ($P < 0.001$, Mann-Whitney *U* test; Fig. 1, Table 2).

TBUT and Fluorescence Staining

The TBUT was reduced from baseline to 6 AM the following day in the SD group ($P = 0.019$, Wilcoxon signed rank test) and recovered to the baseline at 2 PM the following day. At 6 AM the following day, the TBUT was shorter in the SD group compared to the control group ($P < 0.001$, Mann-Whitney *U* test; Fig. 2, Table 2).

Schirmer's Tear Secretion Test

Tear secretion in the SD group was decreased from baseline to 6 AM the following day ($P = 0.005$, Wilcoxon signed rank test), but had recovered to baseline levels by 2 PM the following day. Tear secretion at 6 AM the following day was significantly reduced in the SD group compared to the control group ($P < 0.001$, Mann-Whitney *U* test; Fig. 3, Table 2).

Visual Analog Pain Scale

According to the responses on VAS, pain increased to 6 AM the following day from baseline and 10 PM in the SD group ($P = 0.006$ and $P = 0.004$, respectively, Wilcoxon signed rank test). The VAS score also was higher at 6 AM the following day in the SD group compared to the control group ($P < 0.001$, Mann-Whitney *U* test; Fig. 4, Table 2).

IOP Measurement

No differences in IOP were observed in either the SD or control groups (Fig. 5).

Discussion

Dry eye is a multifactorial disease involving the tear film and ocular surface.[1] The tear film reportedly is affected by a variety

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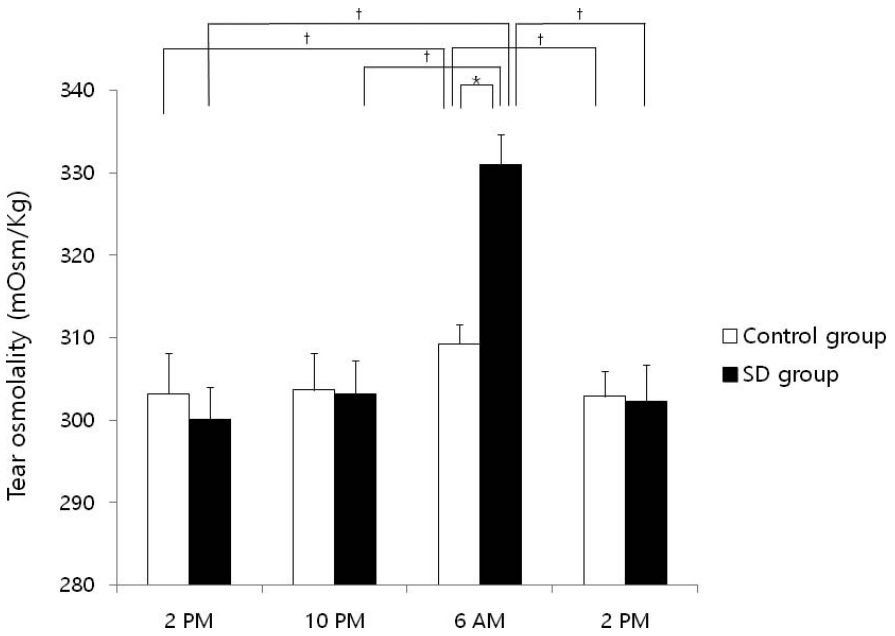

Figure 1. Tear osmolarity in the SD group was increased from baseline to 6 AM the following day ($P = 0.008$, Wilcoxon signed rank test); tear osmolarity also was greater in the SD group compared to the control group at that time ($P < 0.001$, Mann-Whitney U test). *Statistically significant according to Mann-Whitney U test. †Statistically significant by Wilcoxon signed rank test.

of factors.[1,13] For instance, hormonal levels, such as low androgen and high estrogen levels, are risk factors for dry eye.[14] Reduction of tear secretion in Sjögren's syndrome, systemic drug uses, and increasing age,[15] as well as ocular sensory loss in contact lens wear and diabetes also can cause dry eye syndrome.[1] Furthermore, the lipid layer of tear film is disturbed by meibomian gland dysfunction and blepharitis, which is a risk factor for dry eye.[1,16] Recently, dry eye syndrome has been reported to be associated with depression and post-traumatic stress disorder.[17]

Dry eye syndrome has been reported to be more prevalent in females.[1,2] However, only men were enrolled in this study. Sleep deprivation has been reported to show more augmented autonomic neural responses in men compared to women.[18] Nevertheless, if SD could induce dry eye syndrome in men, it likely has a similar effect in women.

Sleep deprivation has been reported to contribute to several disease processes and to reduced longevity,[19] and it also leads to hormonal and neurochemical changes.[7] Sleep disorders, including SD, lead to altered cellular responses,[20] oxidative stress,[21] and increased levels of stress hormones, including norepinephrine and cortisol.[7,22] In this study, we revealed that SD also disturbs tear film. The condition increased tear osmolarity, and hyperosmolarity has been suggested as the primary causative mechanism in dry eye syndrome.[1] Tear osmolarity represents variations in tear dynamics and is an accepted method for diagnosing dry eye syndrome.[1,3]

Table 1. Characteristics of the Subjects at Baseline

	Control Group	SD Group	*P* Value*
Age, y	24.57 ± 1.31	25.25 ± 1.04	0.899
VAS, 0–10	0.56 ± 0.53	1.22 ± 0.67	0.74
TBUT, s	12.7 ± 1.9	13.3 ± 1.0	0.673
Schirmer's test, mm	11.0 ± 2.3	11.2 ± 1.4	0.804
Tear osmolality, mOsm/kg	300.1 ± 3.8	303.2 ± 4.8	0.150
IOP, mm Hg	17.7 ± 1.9	17.7 ± 1.6	0.900

* Mann-Whitney U test.

Sleep deprivation also shortened TBUT and increased the VAS pain score. The TBUT has been reported to represent tear stability.[1] Subjects in the SD group complained of eye discomfort, dryness, and grittiness, and SD was shown to decrease tear secretion by Schirmer's test. Several mechanisms potentially could explain these findings. Tear secretion has been reported to be affected by a variety of factors.[12] First, tears are produced by the lacrimal glands, which are innervated by parasympathetic and sympathetic nerves.[12,23] Sleep deprivation has been reported to heighten the levels of stress hormones, including cortisol, epinephrine, and norepinephrine,[17] and to decrease parasympathetic and increase sympathetic tone.[7,24] Typically, activation of the parasympathetic pathway stimulates tear secretion because parasympathetic fibers are predominant in the lacrimal glands.[12] Second, SD leads to mild activation of the hypothalamic–pituitary–adrenal axis and elevated plasma concentrations of glucocorticoids in humans.[11,25] It also reportedly causes excess diuresis and natriuresis. Although renal water control and arginine vasopressin levels remain unaltered during SD, the circadian rhythm of the renin–angiotensin–aldosterone system hormones is altered significantly.[5] It has been suggested that the underlying mechanism of dehydration following SD could be a reduced nighttime dip in blood pressure and a decrease in renin–angiotensin–aldosterone system levels.[5,6] These alterations in hormone levels and excess dieresis could induce a relatively dehydrated state, which can affect tear secretion.

In this study, water intake was not restricted during the entire experimental period to eliminate the effects of oral water intake. Adequate total water intakes for sedentary adults are on an average between 2 and 2.5 L per day.[26] However, most of the components of fluid balance are controlled by homeostatic mechanisms responding to the state of water in the body.[27] These mechanisms are sensitive and precise, and are activated with deficits or excesses of water amounting to only a few hundred milliliters.[27] Body hydration state is regulated by the renin–angiotensin–aldosterone system and antidiuretic hormone vasopressin.[26] Thus, sleep disorder-

Table 2. Changes in Symptoms, Tear Film, and Ocular Surface After SD

	Baseline, 2 PM	8 h Later, 10 PM	16 h Later, 6 AM	24 h Later, 2 PM
VAS, 0–10*				
Control group	0.56 ± 0.53	0.89 ± 0.60	1.33 ± 0.70†	1.22 ± 0.67
SD group	1.30 ± 0.67	1.40 ± 0.52	4.00 ± 1.15†‡	1.50 ± 0.70
P value	0.063	0.133	< 0.001§	0.548
TBUT, s*				
Control group	12.7 ± 1.9	11.7 ± 1.2	10.4 ± 0.9	11.7 ± 1.4
SD group	13.3 ± 1.0	12.1 ± 1.0	4.0 ± 0.6*	11.7 ± 0.9
P value	0.673	0.502	< 0.001§	0.735
Schirmer's test, mm*				
Control group	11.0 ± 2.3	10.7 ± 1.8	9.8 ± 2.0	10.8 ± 1.8
SD group	11.2 ± 1.4	10.8 ± 1.3	3.6 ± 0.7*	10.8 ± 1.2
P value	0.804	0.869	< 0.001§	0.773
Tear osmolality, mOsm/kg*				
Control group	303.2 ± 4.8	303.7 ± 4.4	309.2 ± 2.4*	302.9 ± 3.1
SD group	300.1 ± 3.8	303.2 ± 4.0	331.0 ± 3.7*	302.2 ± 4.5
P value	0.150	0.711	< 0.001§	0.934
IOP, mm Hg				
Control group	17.7 ± 1.89	16.7 ± 2.0	16.9 ± 1.8	17.4 ± 1.2
SD group	17.7 ± 1.58	16.9 ± 1.9	16.9 ± 1.6	17.4 ± 1.2
P value	0.900	0.740	0.866	0.836
Foveal thickness, μm				
Control group	290.3 ± 10.9	290.1 ± 11.4	290.4 ± 11.1	290.7 ± 11.3
SD group	290.3 ± 11.7	290.8 ± 12.0	289.8 ± 11.5	289.6 ± 11.8
P value	1.0	0.934	0.967	0.805

* $P < 0.05$ by the RM ANOVA test.
† $P < 0.05$ by the Wilcoxon signed-rank test, compared to baseline values of each parameter.
‡ $P < 0.05$ by the Wilcoxon signed-rank test, compared to values at 10 PM.
§ $P < 0.05$ by the Mann-Whitney *U* test, compared SD group with control group.

induced dehydration could affect the renin–angiotensin–aldosterone system.[5,28]

Diurnal variation in tear osmolarity and secretion has been reported.[29–31] Tear secretion and tear osmolarity are reduced upon wakening.[29–31] In this study, tear osmolarity was not hypotonic at 6 AM compared to baseline. This reason for this might be that the subjects' tears were not collected immediately after awakening but 20 minutes after awakening; it has

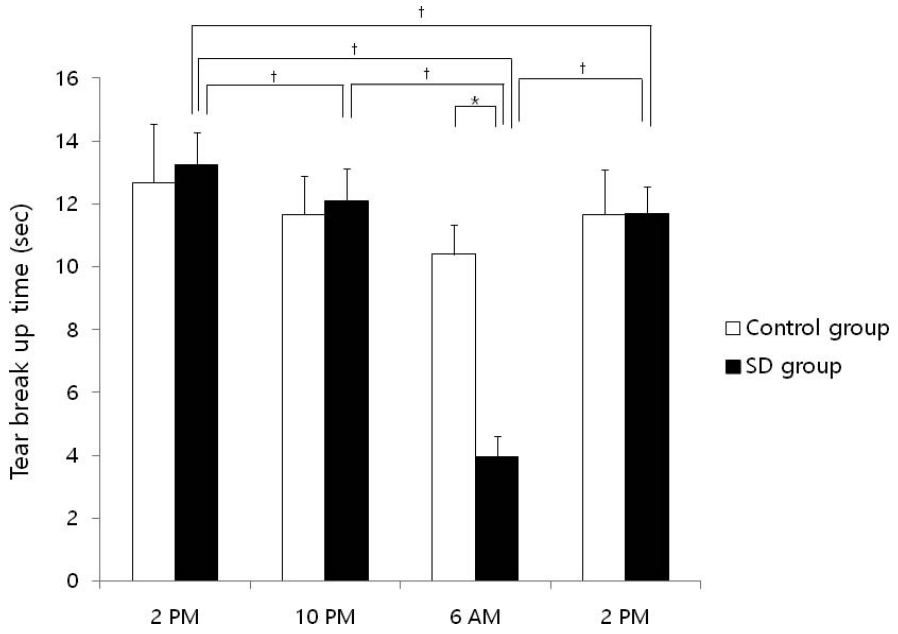

Figure 2. The TBUT in the SD group was reduced from baseline to 6 AM the following day ($P = 0.019$, Wilcoxon signed rank test) and also was shorter in the SD group compared to the control group at that time ($P < 0.001$, Mann-Whitney *U* test). *Statistically significant by Mann-Whitney *U* test. †Statistically significant by Wilcoxon signed rank test.

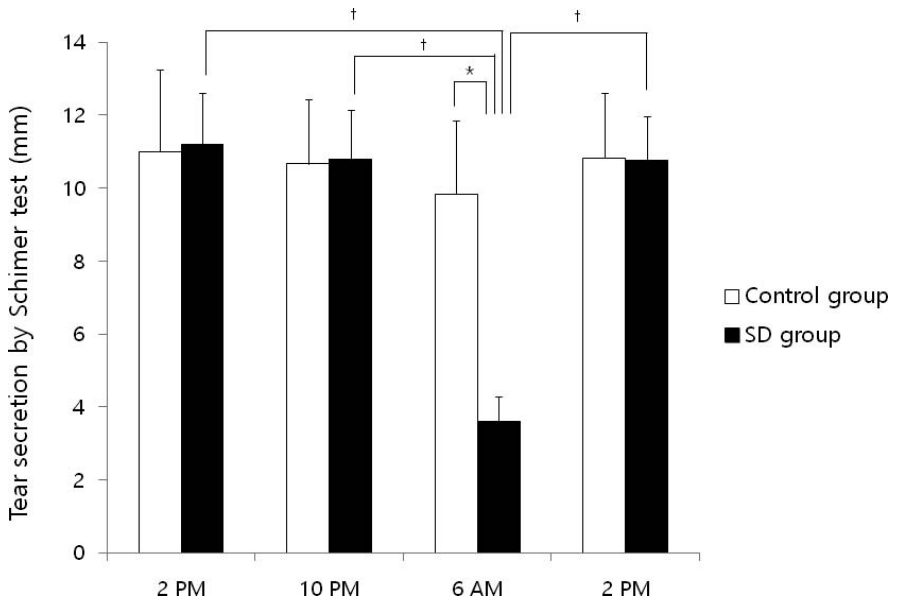

Figure 3. Tear secretion was assessed by Schirmer's test. Tear secretion in the SD group was decreased from baseline to 6 AM the following day ($P = 0.005$, Wilcoxon signed rank test) and also was reduced in the SD group compared to the control group at that time ($P < 0.001$, Mann-Whitney U test). *Statistically significant by Mann-Whitney U test. †Statistically significant by Wilcoxon signed rank test.

been reported that tear osmolarity increased to the baseline levels 20 minutes after waking.[29] All the parameters evaluated in this study returned to normal level 24 hours after SD. These results suggest that SD effects on the ocular surface can be compensated. Firstly, oral intake of food and water during daytime can activate the parasympathetic nerve system, which stimulates the secretion of tear, saliva, and gastric acid.[32] In addition, the serotonin–melatonin imbalance could have normalized over the course of the experiment.[33] Lastly, the renin–angiotensin–aldosterone system might also have normalized.[34]

This study is limited by the small sample size; however, it is a pilot study designed to assess the effect of SD on tear film and the ocular surface. Further studies in a large population are needed to establish associations between sleep disorder and dry eye syndrome and the underlying mechanisms of this relationship.

In conclusion, SD induced tear hyperosmolarity, shortened TBUT, and reduced tear secretion, all of which trigger the development of ocular surface diseases. Thus, SD can exacerbate signs and symptoms in patients with ocular surface diseases.

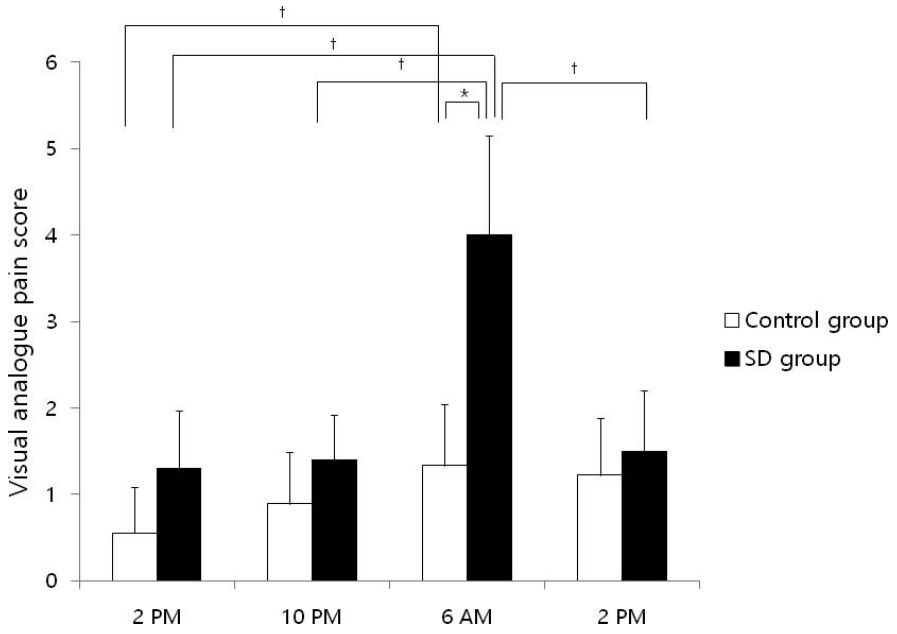

Figure 4. The VAS pain score in the SD group increased from baseline to 6 AM the following day ($P < 0.001$, Wilcoxon rank test) and also was greater in the SD group compared to the control group at that time ($P < 0.001$, Mann-Whitney U test). *Statistically significant by Mann-Whitney U test. †Statistically significant by Wilcoxon signed rank test.

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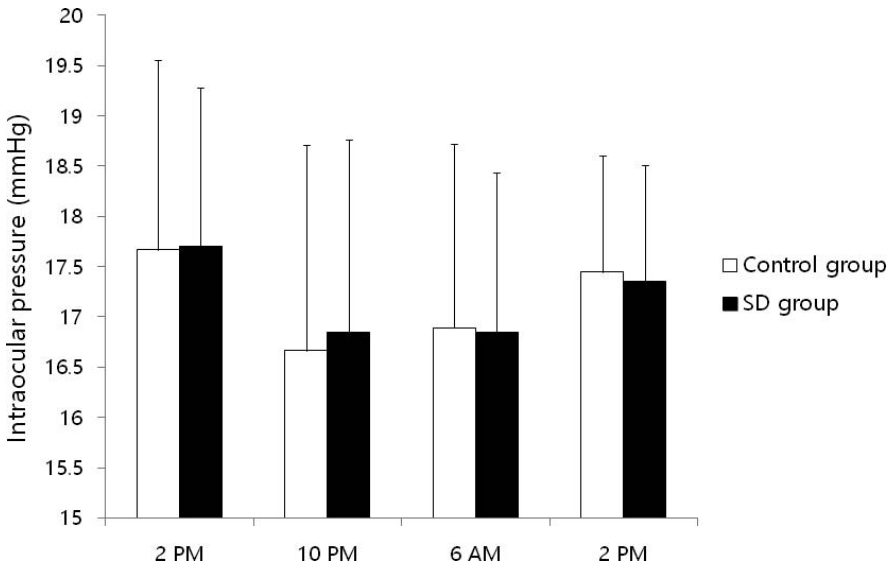

Figure 5. The IOP did not differ in the SD and control groups at any point.

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