

**Influence of sleep restriction on occlusal sensation
in healthy volunteers**

(健全者における睡眠の制限が咬合感覚に及ぼす影響)

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I . Abstract

[Objective]

The aims of the present study were twofold: first, to investigate occlusal contact area of individual teeth during low level tooth clenching, and second to investigate the effect of sleep restriction (SR) on somatosensory sensitivity related to occlusion.

[Materials and methods]

Research 1: Twenty-four healthy participants with complete dentition except for third molars and no known neurological disorders participated in this study. An occlusal contact record was made during three jaw motor tasks (baseline, 20% maximum voluntary contraction (MVC), and 40% MVC) using silicone registration materials. The occlusal contact area (OCA) was calculated by an occlusal analysis device based on the silicone registration materials, defined as level 1 (<150 µm: 0-149 µm), level 2 (<90 µm: 0-89 µm), level 3 (<50 µm: 0-49 µm), level 4 (<30 µm: 0-29 µm), and level 5 (<5 µm: 0-4 µm).

Research 2: This study comprised two experimental sessions (SR and normal sleep: NS) in 12 healthy participants. All participants participated voluntarily in an experimental study involving total SR. In the SR experiment, participants were followed for 3 consecutive days including the 2 nights of sleep. In the NS experiment, all participants were instructed to maintain NS both nights. In all participants tactile detection threshold (TDT), interocclusal detection threshold (IDT), perception of unpleasantness (POU), the Epworth sleepiness scale (ESS) and total sleep time were measured. The measurement points for the TDT, IDT and POU were the lower left first premolar, lower right first premolar, lower left first molar, and lower right first molar.

[Result]

Research 1: At level 1, there was a significant increase from baseline to 40% MVC in the OCA of the right first molar ($P < 0.05$). At level 2, the OCA of the right first molar increased significantly from baseline to 20% MVC, and in the bilateral molars it significantly increased from baseline to 40% MVC ($P < 0.05$). At level 3, the OCAs of the bilateral molars increased significantly from baseline to 20% MVC, and those in right second premolar and bilateral molars increased significantly from baseline to 40% MVC ($P < 0.05$). At level 4, the OCA increased significantly in the right second premolar and bilateral molars from baseline to 20% MVC, and bilaterally in the second premolars and molars from baseline to 40% MVC ($P < 0.05$). At level 5, the OCAs of bilateral premolars and molars increased significantly from baseline to 20% MVC and 40% MVC ($P < 0.05$).

Research 2: Total sleep time on the first night in the SR experiment was significantly shorter than on the second night of the SR experiment and the first night in the NS experiment ($P < 0.05$). ESS values on Day 2 in the SR experiment were significantly higher than on Day 1 and Day 3 in the SR experiment and Day 2 in the NS experiment ($P < 0.05$). In each tooth, the POU was significantly lower on Day 2 in the SR experiment than on Day 1 and Day 3 in the SR experiment and on Day 2 in the NS experiment ($P < 0.05$).

[Conclusion]

The present results suggest that the OCAs in premolar and molar tooth were strongly influenced by tooth clenching intensity, and that SR affects the occlusal sensation related to POU.

II . Introduction

Occlusal dysesthesia (OD) is defined as a persistent uncomfortable sense of intercuspal position after all pulpal, periodontal, muscle, and temporomandibular joint (TMJ) pathologies have been ruled out and a physically obvious bite discrepancy cannot be observed [1]. Comparison of thickness discrimination ability using standard blocks made of stainless steel with thicknesses of 2, 5, and 10 mm between OD patients and normal participants, there is no difference in interdental thickness discrimination ability between the groups. However, their study also investigated the characteristics of OD with a bio-psycho-social approach and suggested that OD patients tend to score higher on psychosomatic distress [2]. A cohort study investigated the relationship between OD and psychosomatic background, management, and treatment outcomes, and they suggested that OD treatment should take into account the underlying psychiatric disorder manifesting such as physical complaints [3]. Although some studies suggested a correlation between OD and psychosomatic factors, there is so far little information to clarify the mechanism of OD. In 2013, although occlusal discomfort syndrome (ODS) was defined as a comprehensive syndrome that is pathologically characterized by discomfort related to occlusion by 'The Japan Prosthodontic Society', this position paper concluded that the final target, yet to be achieved, is to divide this disease into manageable groups and create clinical practice guidelines [4]. However, no clinical diagnostic criteria have been described for evaluating occlusal condition in patients with ODS. To determine clinical diagnostic criteria for ODS, the relationships between occlusal contact area (OCA) in individual teeth need to be established in healthy volunteers.

Sleep disorders are among the most common comorbid problems of chronic pain patients [5,6]. To clarify the effects of sleep restriction (SR) on somatosensory sensitivity, some studies investigated its effects on somatosensory sensitivity of the body. In the effects of SR on thermal pain thresholds and somatosensory thresholds of the skin at the center of the volar forearm, SR produces hyperalgesic changes that cannot be explained by nonspecific alterations in somatosensory functions [7,8]. About the impact of sleep restriction on pain perception of the hands, sleep-restricted participants show reduced attentional modulation of pain stimuli and may thus have difficulties to readily attend to or disengage from pain [9]. Additionally, chronic insufficient sleep may increase vulnerability to chronic pain by altering processes of pain habituation and sensitization [10]. In the orofacial area, SR affects craniofacial muscle sensitivity in healthy humans [11]. However, no studies have addressed the effects of SR on somatosensory sensitivity related to occlusion. To clarify the mechanism of OD, it is essential to investigate the relationship between SR and somatosensory sensitivity related to occlusion.

The aims of the present study were twofold: first, to investigate OCA in individual teeth during low-level tooth clenching, and second to investigate the effect of SR on somatosensory sensitivity related to occlusion.

III. Materials and methods

Research 1: Comparing the occlusal contact area of individual teeth during low-level clenching

Twenty-four healthy participants (12 men, 12 women, mean age \pm standard error of mean 24.3 ± 2.0 years) with complete dentition except for third molars and no known neurological disorders participated in this study. Abnormal stomatognathic

function or bruxism were ruled out by evaluation of dental history using standard questionnaires and self-reports, as well as an oral examination using the research diagnostic criteria for temporomandibular disorder [12]. All participants gave informed consent before the study began. Approval was obtained from the Institutional Ethics Committee (EC05- 015) and the study was conducted in accordance with the guidelines of the Declaration of Helsinki.

Subjects sat upright and relaxed in a dental chair while measurements were taken and the head was supported by a headrest. Before measurements were taken, subjects performed a maximum clench to determine the 100% maximum voluntary contraction (MVC). For the main experiment, subjects were asked to perform three low-intensity tooth clenching tasks (baseline, 20% MVC, and 40% MVC) as previously described [13] for 1 min. To avoid muscle fatigue, a 2 min interval was set between each task. Three jaw motor tasks were performed twice in randomized order. Visual feedback was used to determine 20% and 40% MVC. For baseline measurements, all subjects were instructed to close their mouths and touch the opposing teeth with minimal force. In all measurements, masseter muscle activity was recorded from all subjects.

Disposable bipolar surface EMG electrodes (NM319Y, Nihon Kohden, Tokyo, Japan) were used to record surface EMG activity of the left masseter muscle (LM) and right masseter muscle (RM) in all subjects. The electrodes were positioned parallel to the main direction of the muscle fibers over the lower anterior part of the main muscle belly. This position was determined by palpation about 3 cm superior and anterior to the mandibular angle [14]. The electrodes were positioned 10 mm apart along the central part of the muscle, midway between the anterior and posterior borders and superior and inferior borders of the LM and RM. Visual feedback of muscle activity via the EMG signal

was amplified 500 to 5,000 times and sampled at 1,500 Hz using a muscle balance monitor (GC, Tokyo, Japan). Masseter muscle EMG was recorded during all measurements with a time constant of 0.03 s, sensitivity of 0.5 mV/div, and a sampling frequency of 1 kHz using a multitelemeter system (WEB-5000, Nihon Kohden). EMG signals were analyzed off-line after being transferred to wave analysis software (Powerlab, AD Instruments, Sydney, Australia). EMG activity was initially quantified during each task by calculating the root mean square (RMS) EMG amplitude in each 10 s epoch from both masseter EMG channels in all subjects. The relative ratios in each jaw motor tasks were also calculated from RMS EMG amplitudes of masseter EMG activity.

An occlusal contact record was made during three jaw motor tasks (baseline, 20% MVC, and 40% MVC) using a blue silicone material (Blue Silicone, GC, Tokyo, Japan). The blue silicone material was prepared from an automatic mixing cartridge of silicone materials and injected onto the surfaces of mandibular teeth. The subjects were asked to close their teeth slowly into the maximum intercuspal position and to clench vertically with each jaw motor task for 1 min. The OCA was calculated by an occlusal analysis device (BITEEYE BE-I, GC, Tokyo, Japan) based on the silicone registration materials. Silicone recording materials thicker than 5 mm were unable to calculate the OCA using this occlusal analysis device, therefore silicone recording materials were trimmed to less than 5 mm thick to maintain exact transmittance. As previously described [13], the OCA thicknesses were defined as level 1 (<150 μm : 0-149 μm), level 2 (<90 μm : 0-89 μm), level 3 (<50 μm : 0-49 μm), level 4 (<30 μm : 0-29 μm), and level 5 (<5 μm : 0-4 μm) in the image to calculate the OCA. The OCA in each tooth was divided using a maxillary tooth image on the computer screen and calculated from three jaw motor tasks

according to the five thickness levels. The mean OCA values of each tooth were calculated for each jaw motor task from five different thickness levels of silicone registration material.

One-way analysis of variance was used to analyze the RMS EMG amplitude and the relative ratio of masseter EMG activity. The effects of the task on the OCA of individual teeth were analyzed by the Kruskal Wallis test with multiple comparisons (Bonferroni correction) for each level of silicone registration material thickness. P values < 0.05 were considered statistically significant.

Research 2: Effect of sleep restriction on somatosensory sensitivity including occlusal sensation in the orofacial area

Twelve healthy participants (6 men, 6 women, mean age \pm standard error of the mean 27 ± 2 years) with no neurological and mental disorders participated in this study. Abnormal stomatognathic function or bruxism was ruled out by evaluation of the dental history using standard questionnaires and self-reports, as well as an oral examination using the diagnostic criteria for temporomandibular joint disorder [15]. All participants recruited from staff worked at our dental school and gave their informed consent before the study began. The Institutional Ethics Committee approved the study (EC16-012), and the study was conducted in accordance with the guidelines of the Declaration of Helsinki.

All participants participated in an experimental voluntary total SR study. In a study design, they were invited to sleep as usual, normal sleep (NS) or to restrict their sleep for four nights. Following the SR night, participants were followed for 3 consecutive days including the 2 sleep nights. In NS experiment, all participants were instructed to maintain NS both nights (Fig. 1). During both experiments, participants were instructed

to prohibit excessive intake of caffeine. All participants underwent measurements of tactile detection threshold (TDT) [16,17], interocclusal detection threshold (IDT) [18-22] perception of unpleasantness (POU) [23], and the Epworth Sleepiness Scale (ESS) [24] at six days. The Pittsburgh Sleep Quality Index (PSQI), Generalized Anxiety Disorder 7 (GAD-7), and Patient Health Questionnaire 9 (PHQ-9) were assessed before Day 1 in both experiments. During all nights, actigraphy data were collected using an actiwatch (Actiwatch Spectrum, Philips Respironics, Murrysville, PA, USA) to evaluate whether the subject was asleep or awake based on the acceleration of body and irradiance [25]. Actigraphic data during 30-s epochs were scored as sleep or wake by Actiware software (version 6.0.5, Phillips Respironics), and total sleep time was estimated.

Before all experiments, OCA was measured using silicone materials (Blue Silicone, Tokyo, Japan) in all participants. The measurement points for the OCA were the lower left first premolar (L4), lower right first premolar (R4), lower left first molar (L6), and lower right first molar (R6). An automatic mixing cartridge of silicone materials was used and injected onto the surfaces of lower teeth. The participants were instructed to clench into the intercuspal position with the instruction, 'Please bite down and clench lightly' according to Obara et al. [26]. In this data analysis, the OCA was calculated by an occlusal analysis device (BITEEYE BE-I, GC, Tokyo, Japan) based on the silicone registration materials. The thickness levels for calculating OCA were defined as 0-29 μm in the image to calculate OCA.

The TDT at L4, R4, L6 and R6 was measured by half-cut Semmes-Weinstein Monofilaments (Premier Products, Kent, WA, USA) with 20 different diameters corresponding to 20 target forces (0.3 – 7186.3 mN) on each day in both experiments. Tactile stimulation was applied to the occlusal surface of L4, R4, L6 and R6 in both

experiments. Participants were asked to answer 'YES' or 'NO' regarding perception of the filament on the occlusal surface. TDT was measured three times in L4, R4, L6 and R6 in both experiments using the psychophysical method (method of limits) according to Komiyama et al. [16]. The TDT in each tooth in both experiments was defined as the mean value calculated from the three TDT measurements.

The IDT and POU of L4, R4, L6 and R6 were measured by stacked metal strips (Artus, Englewood, NJ, USA) with multiples of 12- μ m thickness levels (e.g. 12 μ m, 24 μ m, 36 μ m, 48 μ m, 60 μ m, 72 μ m, 84 μ m, 96 μ m, 108 μ m, 120 μ m, 132 μ m, 144 μ m, 156 μ m) on each day in both experiments. The metal strips were placed on the occlusal surface of L4, R4, L6 and R6. Participants were instructed to relax, concentrate, and keep their eyes closed to avoid any external sensory interference. The participants were instructed to clench into the intercuspal position with the instruction, 'Please bite down and clench lightly', according to Obara et al. [26], and bite the metal strips at the intercuspal position for 5 seconds. In the IDT, participants were asked to answer 'YES' or 'NO' regarding the perception of the metal strips when participants bit the metal strips between their teeth. In the POU, participants were asked to answer 'unpleasant' or 'not unpleasant' when they bit the metal strips between their teeth. IDT and POU were measured three times for each tooth in both experiments using the psychophysical method (method of limits). The IDT and POU of each tooth on both experiments were defined as the mean values calculated from the three IDT and POU measurements, respectively.

All data are presented as mean values and standard deviation. Wilcoxon's signed-rank test was used to compare GAD-7, PHQ-9, PSQI, total sleep time, ESS, TDT, IDT, and POU between the SR and NS experiments. Friedman's test was used to

compare ESS, TDT, IDT, and POU within experiments as multiple comparisons (Bonferroni correction). P values < 0.05 were considered significant. All analyses were performed using the SPSS 12.0 package.

IV. Results

Research1 : Comparing the occlusal contact area of individual teeth during low-level clenching

1. EMG measurements

RMS EMG amplitude values at baseline, 20% MVC, and 40% MVC were 0.014 ± 0.002 , 0.037 ± 0.029 , and 0.071 ± 0.069 , respectively, and the relative ratios of masseter EMG activity were $11.1\% \pm 7.3\%$, $21.4\% \pm 8.7\%$, and $37.5\% \pm 13.6\%$, respectively. RMS EMG amplitudes and relative ratios of masseter EMG activity were significantly higher during 40% MVC than those of 20% MVC ($P < 0.05$) and baseline ($P < 0.001$). RMS EMG amplitudes and relative ratios of masseter EMG activity were significantly higher during 20% MVC than at baseline ($P < 0.001$).

2. Occlusal contact area

At level 1, there was a significant increase from baseline to 40% MVC in the OCA in R6 at level 1 ($P < 0.05$) (Fig. 2). At level 2, the OCA in R6 increased significantly from baseline to 20% MVC, and bilateral molars significantly increased from baseline to 40% MVC ($P < 0.05$) (Fig. 3). At level 3, the OCAs in bilateral molars increased significantly from baseline to 20% MVC, and those in R5 and bilateral molars increased significantly from baseline to 40% MVC ($P < 0.05$) (Fig. 4). At level 4, the OCA increased significantly in the R5 and bilateral molars from baseline to 20% MVC, and in bilateral second premolars and molars from baseline to 40% MVC ($P < 0.05$) (Fig. 5). At level 5,

the OCAs in bilateral premolars and molars increased significantly from baseline to 20% MVC and 40% MVC ($P < 0.05$) (Fig. 6). There were no significant differences in OCAs from 20% MVC to 40% MVC at all levels (Figs. 2-6). There were also no significant differences in OCAs of incisors and canines from baseline to 20% and 40% MVC at all levels.

Research2 : Effect of sleep restriction on somatosensory sensitivity including occlusal sensation in the orofacial area

1. Total sleep times, ESS and PSQI

Total sleep times on the first and second nights in the SR experiment were 0.91 ± 0.56 hours and 8.79 ± 1.66 hours. Total sleep times on the first and second nights in the NS experiment were 6.56 ± 0.56 hours and 6.68 ± 0.91 hours. Total sleep time on the first night in the SR experiment was significantly shorter than on the second night in the SR experiment and the first night in the NS experiment ($P < 0.05$) (Fig. 7).

ESS values on Day 1, Day 2, and Day 3 in the SR experiment were 4.69 ± 2.13 , 12.15 ± 6.01 , and 3.46 ± 1.71 , respectively. ESS values on Day 1, Day 2, and Day 3 in the NS experiment were 4.46 ± 2.43 , 4.23 ± 2.86 , and 4.46 ± 2.47 , respectively. ESS values on Day 2 in the SR experiment were significantly higher than on Day 1 and Day 3 in the SR experiment and Day 2 in the NS experiment ($P < 0.05$) (Fig. 8).

PSQI was 3.1 ± 1.85 in the SR experiment and 2.66 ± 1.15 in the NS experiment, with no significant difference ($P = 0.29$).

2. GAD-7 and PHQ-9

GAD-7 was 1.5 ± 1.56 in the SR experiment and 0.83 ± 0.93 in the NS experiment, with no significant difference ($P = 0.16$). PHQ-9 was 2.08 ± 1.72 in the SR

experiment and 1.83 ± 2.03 in the NS experiment, with no significant difference ($P = 0.68$).

3. Occlusal contact area

OCA values at L4, L6, R4, and R6 were $1.30 \pm 1.06 \text{ mm}^2$, $3.13 \pm 2.23 \text{ mm}^2$, $1.58 \pm 1.41 \text{ mm}^2$, and $2.55 \pm 1.38 \text{ mm}^2$, respectively.

4. Tactile detection threshold

Figure 9 shows the comparison of TDT at L4, L6, R4, and R6 between each day in both experiments. There were no significant differences in TDT between each day at each measurement point in both experiments.

5. Interocclusal detection threshold

Figure 10 shows the comparison of IDT at L4, L6, R4, and R6 between each day in both experiments. There were no significant differences in IDT between each day at each measurement point in both experiments.

6. Perception of unpleasantness

Figure 11 shows the comparison of POU at L4, L6, R4, and R6 between each day in both experiments. POU was significantly lower on Day 2 in the SR experiment than on Day 1 and Day 3 in the SR experiment and on Day 2 in the NS experiment ($P < 0.05$).

V. Discussion

Research 1: Comparing the occlusal contact area of individual teeth during low-level clenching

In the present study, premolar and molar OCAs significantly increased at each level from baseline to 20 and 40% MVC, but no significant differences were observed

from 20 to 40% MVC. Clenching intensity did not significantly change the OCA of anterior teeth at all detection levels. Guadsapsri et al. discussed the mechanism by which changes in clenching intensity increase the OCA [27]. Some studies showed that the mandible functions as a class III lever and tension vectors produced by isometric contraction of the jaw-closing muscles lie between the mandibular condyle and the dental arch [28-30]. Mansour et al. suggested that the bite force between the molar teeth increases progressively in a non-linear but monotonic manner as the bite point moves posteriorly [31]. In addition, Kikuchi et al. showed that the relative occlusal force ratio of the second molar increased with increasing clenching levels, while that of the canine decreased [32]. These results suggest that contact distribution is altered in humans by shifts in regional occlusal loads during clenching. Non-rigidity of the bone and periodontal ligament allow minor tooth movement during forceful clenching. The greater the forces between antagonistic teeth, the greater are the tooth movements in the periodontal space, meaning closer intercuspation and reduced space between antagonistic teeth [27]. This may explain the significant increase in OCA in the posterior region.

Research 2: Effect of sleep restriction on somatosensory sensitivity including occlusal sensation in the orofacial area

This study investigated the effects of sleep restriction on somatosensory sensitivity related to occlusion. Although there were no significant differences in TDT and IDT between each day at each measurement point in both experiments, POU on Day 2 in the SR experiment was significantly lower than on Day 1 and Day 3 in the SR experiment and Day 2 in the NS experiment.

Some functional near-infrared spectroscopy studies investigated the

relationship between brain activity and an uncomfortable sense of intercuspal position and suggested a correlation between brain activity at the prefrontal cortex and OD [33-35]. On the other hand, some studies found a correlation between SR and brain activity at the prefrontal cortex in humans [36,37]. In the transportation of the signal from body parts to central nervous system, reduced filtering capacities or further factors affecting the strength of the signal or the capacity of sensory filters [38]. The prefrontal cortex exerts top-down influences on several aspects of higher-order cognition by functioning as a filtering mechanism that biases bottom-up sensory information toward a response [39]. Since our present study showed that POU on Day 2 in the SR experiment was significantly lower than on Day 1 and Day 3 in the SR experiment and Day 2 in the NS experiment, our present results may suggest that the effect of SR on occlusal sensation is caused not by the peripheral nervous system, but by the central nervous system.

In this study, mean TDT values were from 23.7 mN to 86.7 mN. A previous study demonstrated that the mean TDT from the first premolar to the second molar was from 32.4 mN to 97.1 mN [17]. The present result was similar as the previous ones. On the other hand, other study investigated the effect of SR on somatosensory sensitivity of the hands using QST, and they suggested that SR significantly affected pain thresholds but not detection thresholds [40]. The present result suggests that SR also had no effect on TDT in the orofacial area.

In the present study, the mean IDT values were from 22.33 μm to 33.5 μm . Some studies investigated the IDT using several devices, and IDT during tooth bite was from 9 to 50 μm [18-22]. The variability of IDT in these studies depended on several factors (i.e. device, experimental design, etc.). The present results demonstrated that there were no significant differences in IDT between each day at each measurement

point in both experiments. A previous study suggested that, for an interincisor distance of 5 mm or more, non-periodontal receptors such as muscular or articular receptors play a predominant role [41]. The present results suggest that SR did not affect somatosensory sensitivity related to periodontal receptors.

There were no significant differences in GAD-7, and PHQ-9, and PSQI between the SR and NS experiments. Our present study demonstrated that washout period of our experimental design appropriated. However, some studies showed that SR affects psychological factors [42-44]. Since our present study did not measure GAD-7 and PHQ-9 in each day at each measurement point in both experiments, further studies are needed to investigate the relationship between psychological factors related to change of sleep condition and somatosensory sensitivity in the orofacial area.

VI. Conclusion

The present results suggest that the OCAs in premolar and molar tooth were strongly influenced by tooth clenching intensity, and that SR affects the occlusal sensation related to POU.

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VIII. Table and Figures

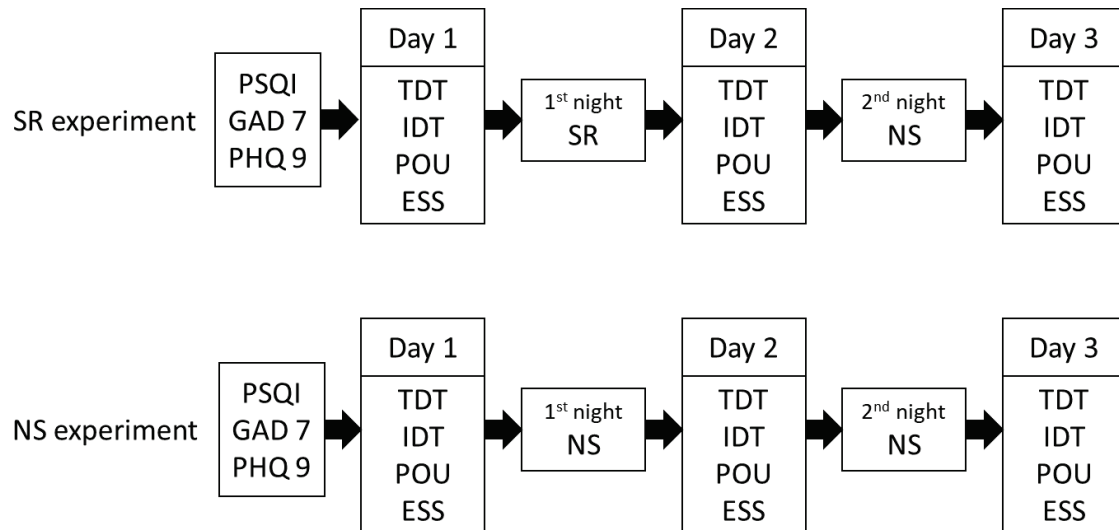


Figure 1. Diagram of study design.

SR, sleep restriction; NS, normal sleep; TDT, tactile detection threshold; IDT, Interocclusal detection threshold; POU, perception of unpleasantness; ESS, Epworth Sleepiness Scale; PSQI, Pittsburgh Sleep Quality index; GAD 7, Generalized Anxiety Disorder 7; PHQ 9, Patient Health Questionnaire 9

Thickness level 1

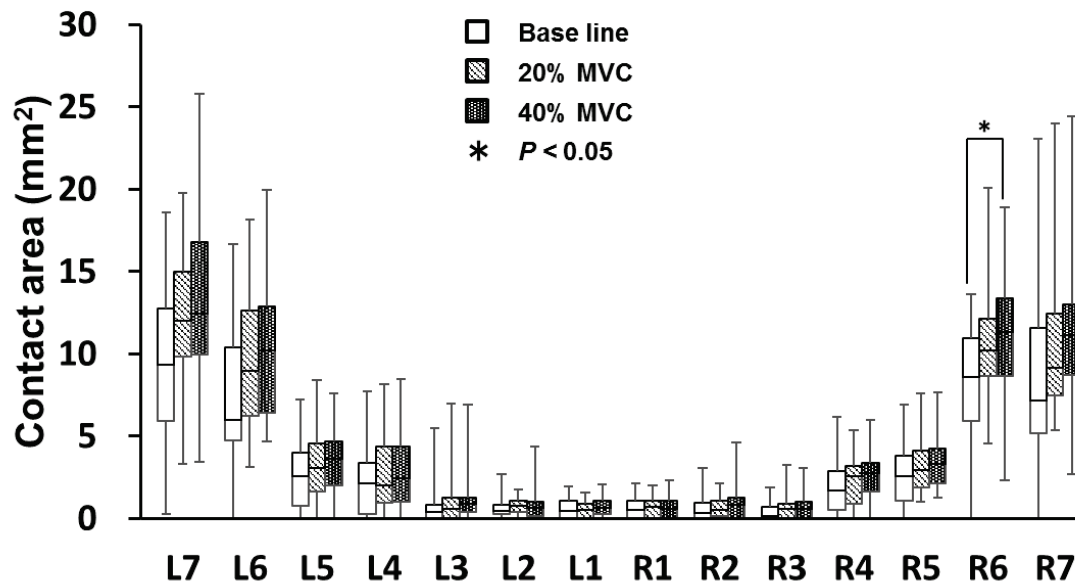


Figure 2. Changes in the occlusal contact area of individual teeth at thickness level 1.

* $P < 0.05$: Kruskal Wallis test with multiple comparisons (Bonferroni correction).

L7, left side second molar; L6, left side first molar; L5, left side second premolar; L4, left side first premolar; L3, left side canine; L2, left side lateral incisor; L1, left side central incisor; R1, right side central incisor; R2, right side lateral incisor; R3, right side canine; R4, right side first premolar; R5, right side second premolar; R6, right side first molar; R7, right side second molar; MVC, maximum voluntary contraction.

Thickness level 2

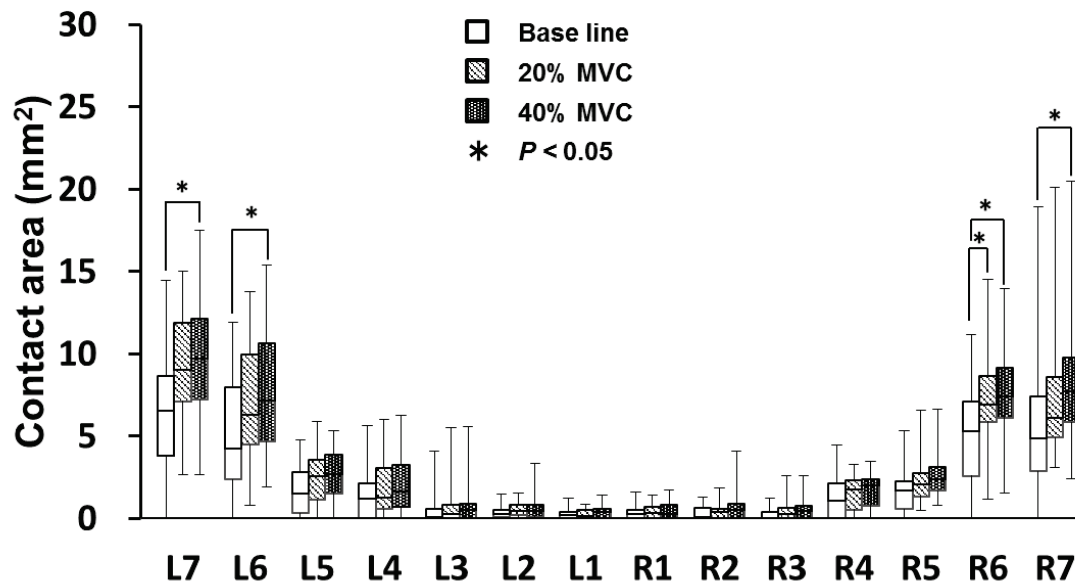


Figure 3. Changes in the occlusal contact area of individual teeth at thickness level 2.

* $P < 0.05$: Kruskal Wallis test with multiple comparisons (Bonferroni correction).

L7, left side second molar; L6, left side first molar; L5, left side second premolar; L4, left side first premolar; L3, left side canine; L2, left side lateral incisor; L1, left side central incisor; R1, right side central incisor; R2, right side lateral incisor; R3, right side canine; R4, right side first premolar; R5, right side second premolar; R6, right side first molar; R7, right side second molar; MVC, maximum voluntary contraction.

Thickness level 3

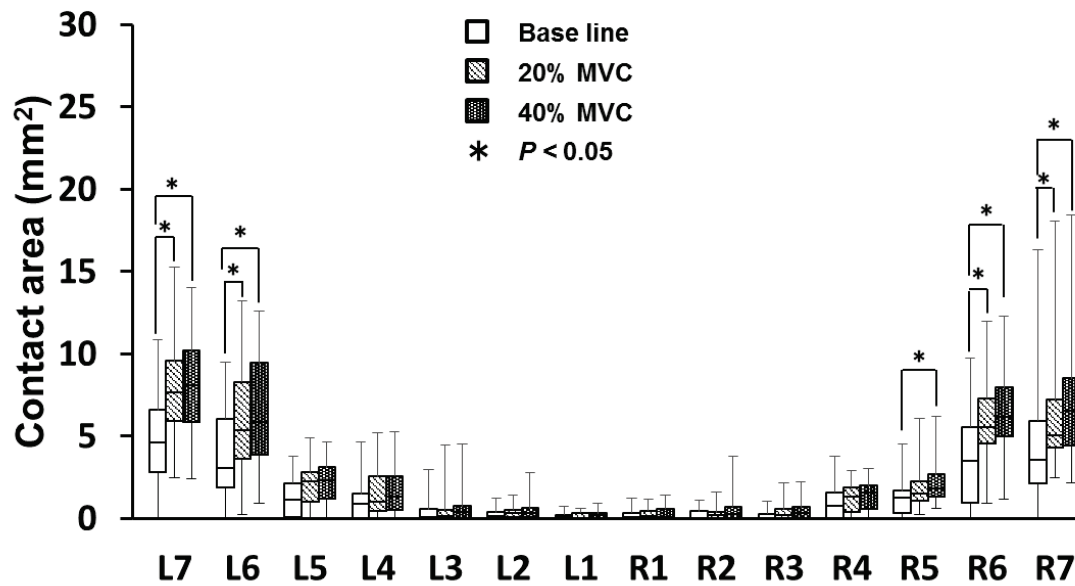


Figure 4. Changes in the occlusal contact area of individual teeth at thickness level 3.

* $P < 0.05$: Kruskal Wallis test with multiple comparisons (Bonferroni correction).

L7, left side second molar; L6, left side first molar; L5, left side second premolar; L4, left side first premolar; L3, left side canine; L2, left side lateral incisor; L1, left side central incisor; R1, right side central incisor; R2, right side lateral incisor; R3, right side canine; R4, right side first premolar; R5, right side second premolar; R6, right side first molar; R7, right side second molar; MVC, maximum voluntary contraction.

Thickness level 4

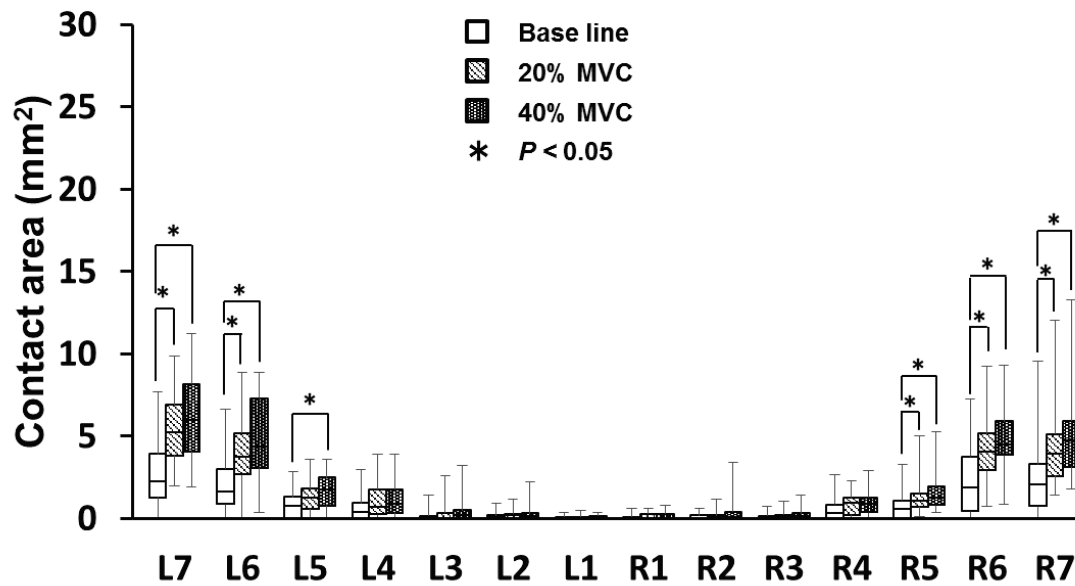


Figure 5. Changes in the occlusal contact area of individual teeth at thickness level 4.

* $P < 0.05$: Kruskal Wallis test with multiple comparisons (Bonferroni correction).

L7, left side second molar; L6, left side first molar; L5, left side second premolar; L4, left side first premolar; L3, left side canine; L2, left side lateral incisor; L1, left side central incisor; R1, right side central incisor; R2, right side lateral incisor; R3, right side canine; R4, right side first premolar; R5, right side second premolar; R6, right side first molar; R7, right side second molar; MVC, maximum voluntary contraction.

Thickness level 5

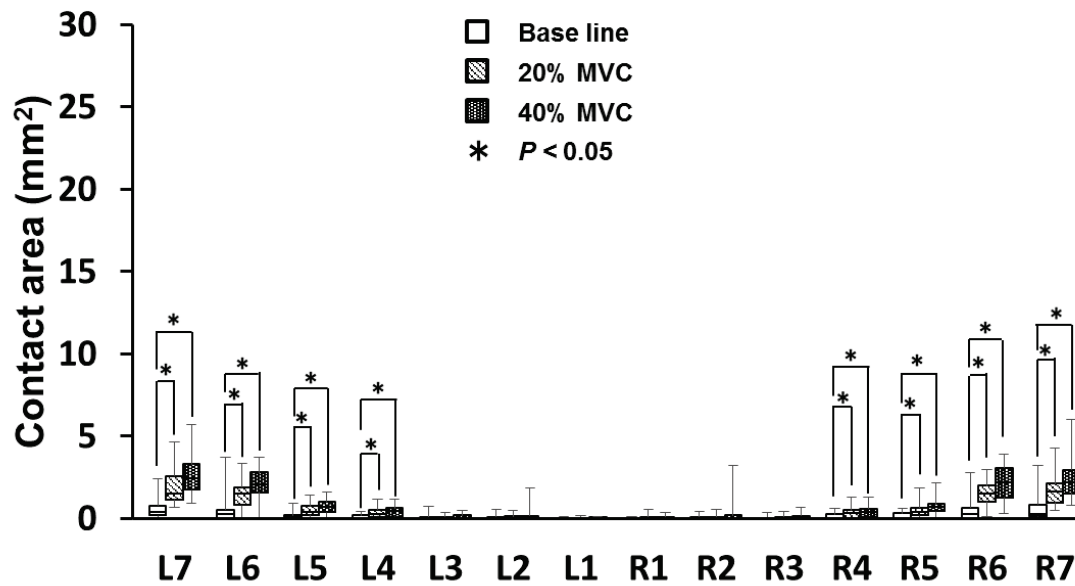


Figure 6. Changes in the occlusal contact area of individual teeth at thickness level 5.

* $P < 0.05$: Kruskal Wallis test with multiple comparisons (Bonferroni correction).

L7, left side second molar; L6, left side first molar; L5, left side second premolar; L4, left side first premolar; L3, left side canine; L2, left side lateral incisor; L1, left side central incisor; R1, right side central incisor; R2, right side lateral incisor; R3, right side canine; R4, right side first premolar; R5, right side second premolar; R6, right side first molar; R7, right side second molar; MVC, maximum voluntary contraction.

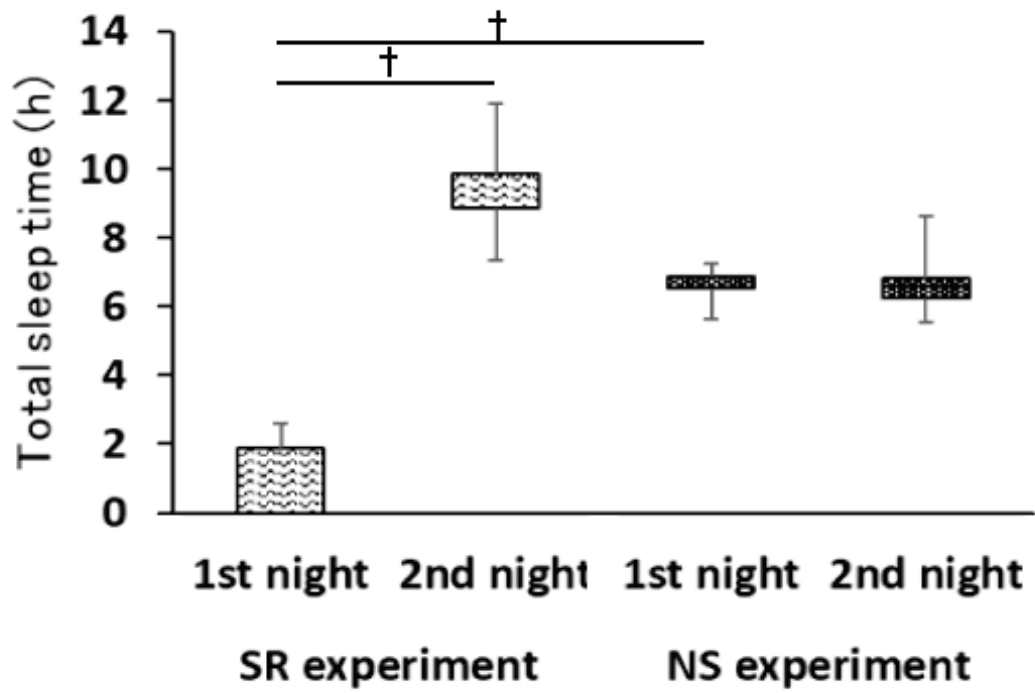


Figure 7. Comparison of total sleep time on each night in both experiments.

† P < 0.05, Wilcoxon signed rank test.

SR, sleep restriction; NS, normal sleep.

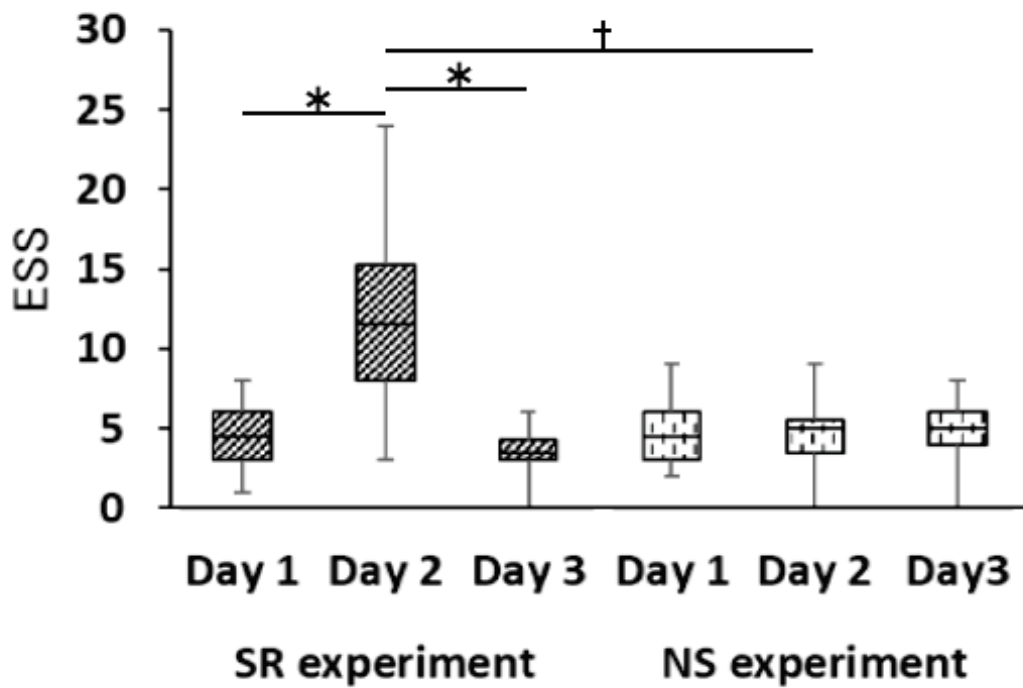


Figure 8. Comparison of ESS on each day in both experiments.

† $P < 0.05$, Wilcoxon signed rank test.

* $P < 0.05$: Friedman's test with multiple comparisons (Bonferroni correction).

ESS, Epworth Sleepiness Scale; SR, sleep restriction; NS, normal sleep.

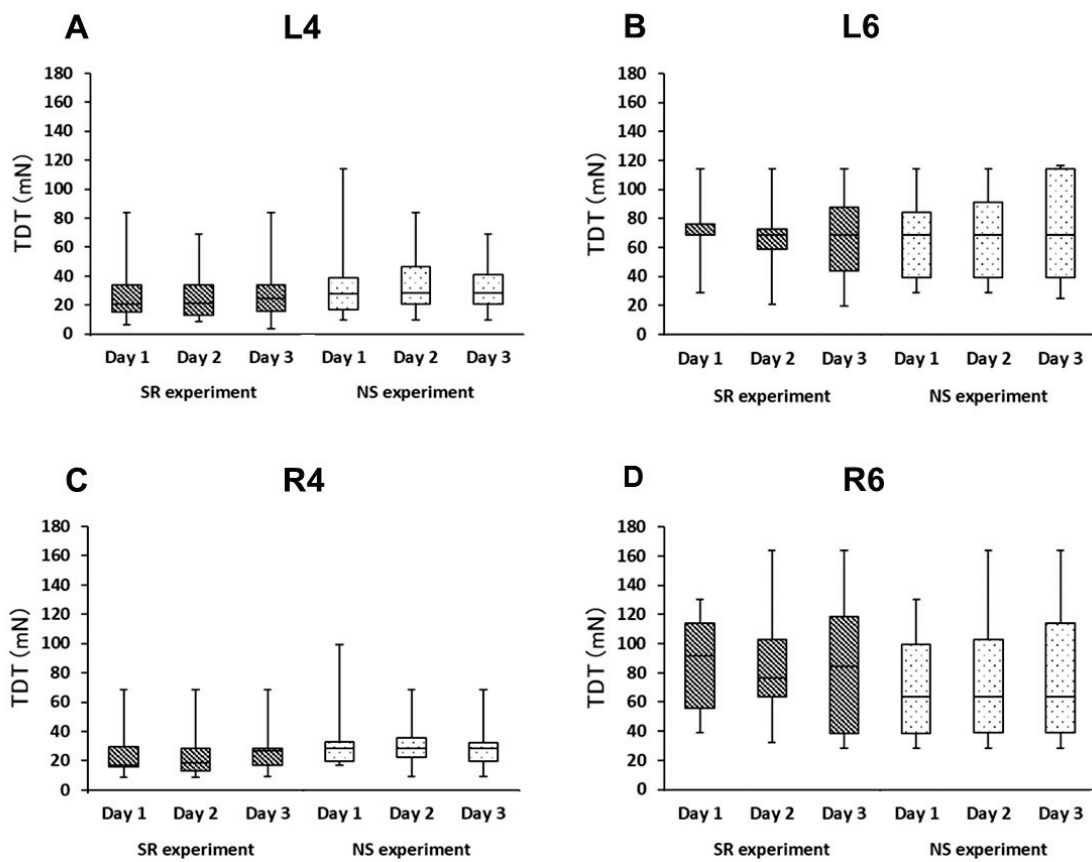


Figure 9. Comparison of TDT at L4 (A), L6 (B), R4 (C), R6 (D) on each day in both experiments.

TDT, tactile detection threshold; L4, lower left first premolar; L6, lower left first molar; R4, lower right first premolar; R6, lower right first molar; SR, sleep restriction; NS, normal sleep.

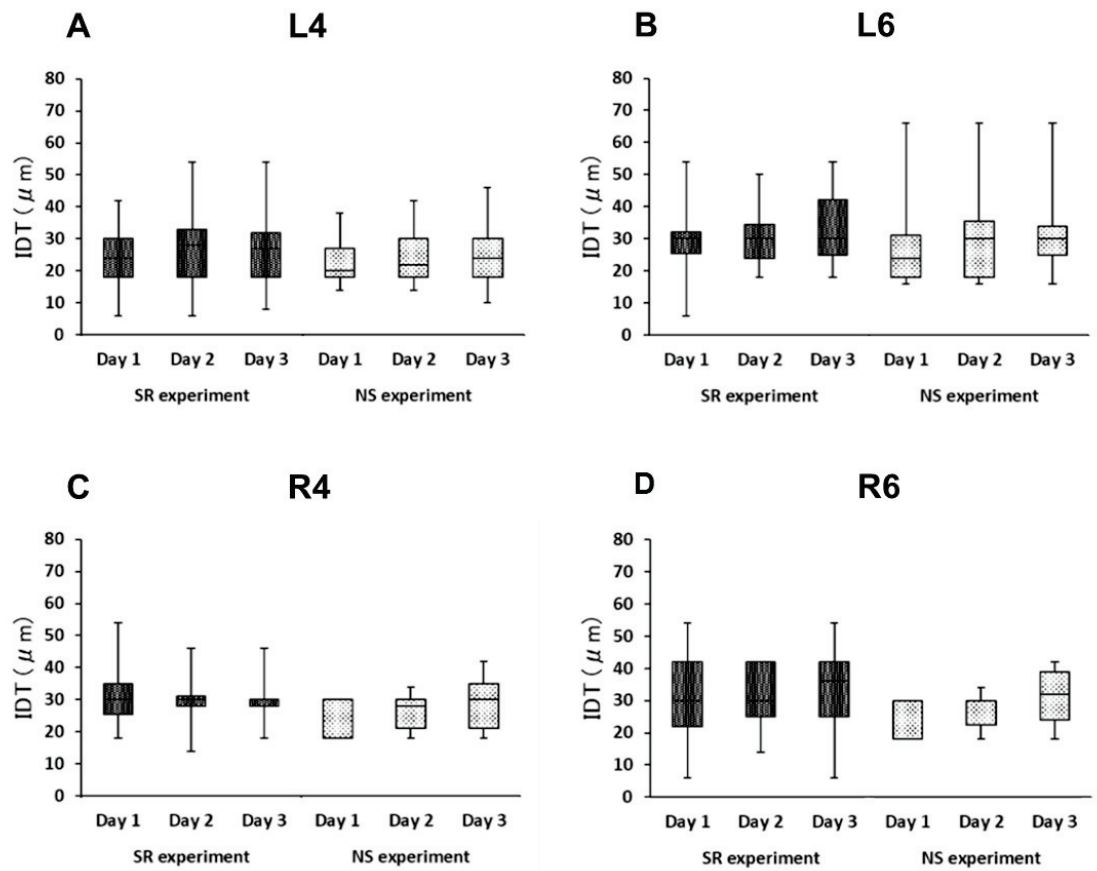


Figure 10. Comparison of IDT at L4 (A), L6 (B), R4 (C), R6 (D) on each day in both experiments.

IDT, Interocclusal detection threshold; L4, lower left first premolar; L6, lower left first molar; R4, lower right first premolar; R6, lower right first molar; SR, sleep restriction; NS, normal sleep.

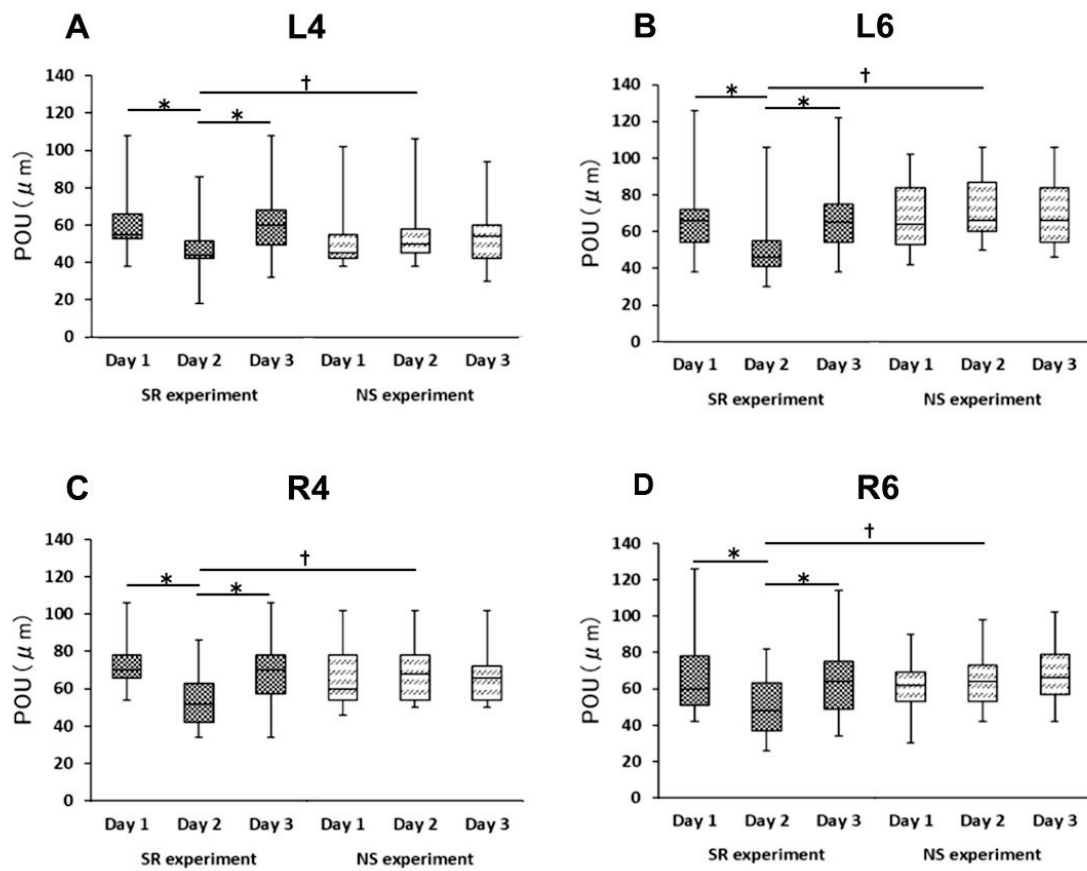


Figure 11. Comparison of POU at L4 (A), L6 (B), R4 (C), R6 (D) on each day in both experiments.

† P < 0.05, Wilcoxon signed rank test.

* P < 0.05: Friedman's test with multiple comparisons (Bonferroni correction).

POU, perception of unpleasantness; L4, lower left first premolar; L6, lower left first molar; R4, lower right first premolar; R6, lower right first molar; SR, sleep restriction; NS, normal sleep.