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Original article

Effects of mats with "A Distinctive 4-Layer 3-Dimensional Structure" on "sleep quality"-related stress and salivary oxytocin: An open-label study

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Abstract

***Objectives*:** "Quality of sleep" plays an important role in maintaining the body's homeostasis. Deterioration of sleep quality is related to stress and can induce various stresses in the body. The present study, an open-label trial, aimed to verify the effects of a test product, a bed mattress with a distinctive 4-layer, 3-dimensional structure, on sleep quality and the secretion of salivary oxytocin (OT).

***Methods*:** Potential research participants included 27 adults (men and postmenopausal women aged 45-64) who reported complaints about sleep quality. From the potential participants, 12 subjects, whose scores of the Pittsburgh Sleep Quality Index (PSQI-J) were six or higher, were selected (age : 54.0 ± 1.8). Test product mattresses from Nishikawa Co., Ltd. (Tokyo) were used for one week, and alternations in physical information were examined in an open trial. The open trial focused on subjective and objective symptom assessments such as Obstructive Sleep Apnea (OSA), Profile of Mood States (POMS2), and State-Trait Anxiety Inventory (STAI), as well as assessments of salivary OT and metabolites of urinary catecholamine were included. The present study was conducted with the approval of an ethics committee.

***Results*:** Regarding subjective and objective symptoms, the following showed significant improvement: in OSA, factor 1 (sleepiness upon rising), factor 2 (initiation and maintenance of sleep), factor 4 (refreshing feeling), and factor 5 (sleep duration). Scores in POM2 for feeling conditions, and STAI for trait anxiety significantly improved. No significant differences were observed in salivary and urinary OT. However, the results of the analysis separated by sex showed that salivary OT levels at wake-up time for women were significantly reduced (2.21 ± 0.25 pg/mL → 1.83 ± 0.16 pg/mL, $p = 0.028$) and the OT level at bedtime was higher than OT level at wake-up time ($p = 0.035$). Regarding catecholamine metabolites, vanillylmandelic acid (VMA) significantly increased in both total of men and women ($p = 0.042$) as well as women alone ($p = 0.035$).

***Conclusion*:** The conditions of subjects before the intervention suggested that OT secretion at wake-up time was accelerated due to responses to stress related sleep quality deterioration. However, it suggested that the usage of the test product could mitigate stresses so that OT, a stress-regulating hormone, decreased. Simultaneously, it was considered that effective stimuli recovered OT secretion capacity and OT secretion at bedtime improved. Improvement in "sleep quality" is important for alleviating mental and physiological stress and maintaining homeostasis in OT secretion.

***KEY WORDS*:** mattress with a "Distinctive 4-Layer 3-Dimensional Structure," sleep quality, oxytocin, norepinephrine, vanillylmandelic acid

Introduction

Oxytocin (OT) is a type of neuropeptide which is produced in the nerve cells of the hypothalamus in the brain. OT is sitimulated by responses toward diverse stresses and stimuli of relations such as sense of touch, vision, smell, and hearing, and released into the brain and bloodstream. This hormone is related to childbirth, breastfeeding and child rearing, promoting contraction of the uterus during childbirth, supporting uterine recovery during puerperium, and facilitating breast milk secretion. Moreover, OT plays a significant role in expressions of love during child rearing [1,2)] and in the establishment of social bonds [3)].

OT is deeply involved in stress responses mentally and

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physically[4]. In the early stages of stress stimulations, the sympathetic nerve is predominant and tachycardia is presented. When the parasympathetic nerve becomes predominant due to OT secretion, tachycardia is alleviated. This means that OT enhances the immune system and plays a role in the maintenance of homeostasis.

Rest and sleep are important for addressing mental and physiological stresses. However, there have not been sufficient findings on how OT is related to sleep quality. We have conducted several clinical trials with subjects who had troubles sleeping[5-10] due to inappropriate bedding to examine how improvements in sleep quality affected impacts on the body. It is possible that deteriorated sleep quality could have impacts on diverse functions of the body and cause trouble in recovering from damages induced by stresses.

We conducted investigations with male and female subjects who had mild sleep disorders, employing a mattress with a "distinctive 4-layer 3-demensional structure." Alternations in salivary OT were assessed, and relationships among stresses, sleep, and OT secretion were analyzed.

Methodss

Research participants

Twenty-seven adults (men and postmenopausal women; ages 45～64) were recruited as potential research participants who submitted written informed consent. Applicants underwent screening such as background examinations, body measurements, Pittsburgh sleep quality index (PSQI-J), Profile of Mood States 2nd Edition; Japanese version (POMS2®) (all items version), State-Trait Anxiety Inventory-Form JYZ (STAI), and doctor's interview. Among potential subjects, 12 subjects were selected for this trial, who satisfied the inclusion criteria and did not meet the exclusion criteria, and had scores of six or higher in PSQI-J, with references to POMS2®, and STAI.

Key inclusion criteria were as follows:

1) Males and premenopausal females aged between 45 and 64 at the time of informed consent
2) Individuals who are healthy and have no chronic physical disease, including skin disease
3) Individuals who are aware of mild sleep disorders such as waking up in the middle of the night (midway awakening), waking up early in the morning (early morning awakening), and not achieving deep sleep (deep sleep disorder)
4) Individuals who work daytime for three-five days a week and have a day off for Saturday and Sunday
5) Individuals who sleep for over four hours, regularly going to bed (lights-out) and waking up
6) Individuals who sleep alone
7) Individuals who do not have a habit of drinking alcohol
8) Individuals who are fully informed regarding the purpose and contents of the test, have the ability to consent, voluntarily apply for participation with full understanding, and agree to participate in the test with written informed consent
9) Individuals who can come to the designated venue on the designated date to undergo the examination
10) Individuals judged appropriate for this study by a responsible doctor

Key exclusion criteria were as follows:

1) Individuals who have contracted diseases and receive medical treatment
2) Individuals who are under treatment for or have a history of mental disorders, sleep disorders, hypertension, diabetes, lipid metabolism abnormality, or other serious disorders
3) Individuals who have a history of and/or are currently experiencing serious diseases (uterine disease, hepatic, renal, cardiovascular, respiratory, hematologic, etc.)
4) Individuals who have a serious history and/or current digestive disease and comorbidities
5) Individuals receiving hormone replacement therapy
6) Individuals suspected of, receiving treatment for, or with a history of sleep apnea syndrome (SAS)
7) Individuals who have or are suspected to have night urination or overactive bladder
8) Individuals who are receiving/received medical drug treatment for the past month except for temporary relief medication for headaches, menstrual pain, common cold, etc.
9) Individuals whose body mass index (BMI) is 30.0 kg/m^2 or more
10) Individuals who have a habit to use functional foods and/or are planning to use those foods during test periods (it is accepted to stop usage for the trial by enrollment)
11) Individuals who donated 200 mL of blood in the past month or more than 400 mL within 3 months
12) Individuals who have possible changes of life style during test periods and with possibility of being unable to use the test product mat due to travel, etc.
13) Individuals who are participating and/or had participated in other clinical studies within the last 3 months
14) Individuals who are judged as not appropriate to this study by a responsible doctor

Trial design

The present study was an open-label uncontrolled trial.

During observational period I, subjects slept on their own daily used bedding, and on the test product (a mattress) during observational period II. The test product was a bed mattress with a distinctive 4-layer 3-dimensional structure, "AiR SX" (Nishikawa Co. Ltd., Tokyo, Japan). The size of the test product was a single size (9 × 97 × 200 cm). The mattresses with the specified sheets were provided by Nishikawa.

Both observational periods lasted seven days. Surveys of effects on the body were examined at the time of five, six, and seven days after the commencement of usage. Moreover, at the time of completion of observational period II, examinations were conducted during the subjects' visit, where the safety of the test product usage was confirmed via physical examination and a medical interview by a doctor. Primary efficacy outcome items consisted of clinical biomarkers and quality of life (QOL)-related biomarkers. The primary safety evaluation items included blood pressure/pulse measurement, weight/body fat percentage/BMI, doctor's interview/judgment on adverse events, and the subjects' daily records. Prior examinations of screening were subjects' background and measurement of subjects' body height. The trial period was October and November of 2023.

Assessment items

Subjective symptoms (sleep)

Qualities of sleep were assessed by the obstructive sleep apnea (OSA) sleep inventory MA version. This is a four-level scale to provide mental assessments on inwardly examining sleep at wake-up time. Subjects answered the questionnaire with scores regarding time to go to bed, time to wake up, and sleep duration. Data results were totaled separately by five factors: factor 1 (sleepiness upon rising), factor 2 (initiation and maintenance of sleep), factor 4 (refreshing feeling), and factor 5 (sleep length).

Subjective symptoms (anxiety and stress)

Subjective symptoms of anxiety and stress were assessed by POMS2® and STAI.

Subjects answered 65 questions of POMS2® with five-rating responses from "never" to "very frequently." Seven scales were "Anger-Hostility," "Confusion-Bewilderment," "Depression-Dejection," "Fatigue-Inertia," "Tension-Anxiety," "Vigor-Activity," and "Friendship." "TMD scores" represented negative feelings in an integrated manner. The aggregation method, based on the instruction manual of POMS2® Japanese version, was used to calculate T scores for all scales.

Subjects provided answers to STAI as follows: twenty items of state anxiety employed a four-point scale from "never corresponding" to "highly corresponding," and twenty items of trait anxiety employed a five-point scale from "almost never" to "almost always." Data results were evaluated as an aggregation method of profiling by sex: Trait Anxiety total scale scores, and P items and A items respectively, State Anxiety and Trait Anxiety. Data were expressed in five levels, T scores and percentiles.

Physical measurement

Body height, weight, somatic fat rate, body fat quantity, fat removal quantity, muscle mass, BMI, contraction and diastolic blood pressure, and pulse rate were measured in the body measurement. A multi frequency segmental body composition analyzer was employed for the examination of body composition (MC-180: Tanita corporation, Tokyo).

Salivary oxytocin (OT) level examination

Samples of salivary OT were collected at subjects' residences in a specified method at bedtime on the 6 th day and wake-up time on the 7 th day respectively, during observational periods I and II. The samples (saliva) were frozen, and the frozen samples were collected when subjects visited the institution, and OT measurements were performed. However, delipidation of samples was not performed.

Urine examination

Samples of first-catch urine (urine stored at night) were collected at subjects' residences in a specified method on the 5th, 6th, and the 7th days respectively in observational periods I and II. The samples (urine) were frozen, and the frozen samples were collected during the subjects' visit. Measurements were performed for OT, melatonin and six components of urine stress marker analysis (dopamine, serotonin, γ-aminobutyric acid [GABA], 5-hydroxyindoleacetic acid, homovanillic acid, and vanillylmandelic acid). The calculation method uses the mean values from three days.

Biological samples were collected in Ueno-Asagao Clinic (Head: Dr. Takahiro Ono, Tokyo) and evaluated by LSI Medience Corporation (Tokyo).

Statistical analysis

Values of examination results were calculated in the ground total sheet, using Microsoft Office Excel 2016 (Microsoft Corp.). As fundamental statistics, the mean values, standard deviation, and standard error were calculated. Values that were used for statistical analysis were measured values and alternation quantity from the value before the usage. Each examination item that was obtained as category data was calculated based on the time of examination. Statistical analysis was performed using appropriate statistical software such as SAS (SAS 9.4) or SPSS (Statistics 26). For all tests, the significance level was 5% and the trend was 10% in two-tailed tests.

Statistical data were analyzed in a paired t-test with comparison between the points during non-usage and during usage. Scores that were obtained in the questionnaire surveys with a Likert scale were treated as non-parametric, and a Wilcoxon signed-rank sum test was conducted for comparison among groups

Ethics review

The present study was conducted with an ethical approval obtained from the committee of "medical research involving human subjects" of a general incorporated association: Society for Glycative Stress Research (October 25, 2023, GSE #2023-003). An informed consent process was provided to potential research participants prior to initiating the research, and the obtained written consents represented the voluntary agreement of research participants. Pre-registration for a clinical trial to University Hospital Medical Information Network, UMIN Clinical Trials Registry (UMIN-CTR) was conducted (registered number: UMIN #000052607). Furthermore, considering the social situation of COVID-19 infections and their impacts, we conducted the present study with the exercise of extreme caution in accordance with the guidelines for infection prevention of COVID-19 by the medical institution for the trial. Only in the case where study directors of the entrusted, the principal investigator, and the ethical committee of medical research judged that the trial was able to be conducted safely.

Results

The present study of an open-label trial with subjects of men and postmenopausal women between 45 and 64 years of age at the time of the obtainment of consent was to examine effects of the test product, a mattress with a distinctive 4-layer 3-dimensional structure, "AiR SX". The dynamic state of OT in the usage of the mat was examined to verify

that improvements in sleep quality were related to diverse mental and physiological symptoms from an endocrinological perspective.

Recruitment procedures are as follows: Telephone interview surveys were conducted with registrants in clinical trial volunteer associations for subject recruitment. Twenty-seven applicants underwent screening when they visited the institution. Among the potential subjects, 12 adults were selected for this trial. They met the inclusion criteria and did not meet the exclusion criteria, were deemed appropriate by a responsible doctor, and had a score of 6 or higher in PSQI-J. The selection was based on the following references:

State-Trait Anxiety Inventory-Form JYZ and POMS2® (all items version)

Additionally, the subjects' backgrounds were considered in the selection process.

During observational period I, subjects slept on their own regularly used bedding, and during observational period II they slept on the test product (a mattress). Surveys of the effects on the body were conducted five, six, and seven days after the commencement of usage. Moreover, upon the completion of observational period II, examinations were conducted during the subjects' visit, where the safety of the test product usage was confirmed through physical examinations and medical interviews by a doctor. The primary efficacy outcome items included clinical biomarkers and QOL-related biomarkers. The primary safety outcomes were blood pressure and pulse measurements, weight, body fat percentage, BMI, the doctor's assessment of adverse events, and subjects' daily records. Prior examinations involved evaluating the subjects` backgrounds and measuring their body height. The present trial started with 12 subjects and was completed with 12 subjects with no cases of withdrawal or termination.

< Primary efficacy outcome items and primary safety outcome items >

All 12 subjects enrolled in the present trial were included in the intention-to-analysis (ITT,) serving as the targets for both primary efficacy and primary safety outcomes. The targets of the ITT analysis targets were 12 subjects (six men and six women) with an average age of 54.0 ± 1.8 (men: 49.7 ± 1.2, women: 58.3 ± 2.4).

Primary efficacy outcome

Subjective and objective symptoms (Table 1)

Examination data of the OSA Sleep Inventory MA version confirmed that subjective symptoms of sleep self-reflection significantly improved. Compared to the period when the test product mattress was not used, significant increases were observed in factor 1 (sleepiness upon rising)

Table 1. Results of OSA, POMS2 and STAI.

	Item	Before Mean SD	After 1 week Mean SD	p value
OSA	Factor 1: sleepiness on rising	40.9 ± 6.8	46.5 ± 5.4	0.003
	Factor 2: initiation and maintenance of sleep	38.3 ± 5.7	47.3 ± 6.0	0.004
	Factor 3; freaquent dreaming	49.4 ± 8.4	49.9 ± 8.1	0.944
	Factor 4: refreshing	41.5 ± 4.3	50.4 ± 5.8	0.003
	Factor 5: sleep length	42.2 ± 8.1	45.9 ± 6.7	0.034
POMS2 (Japan version)	AH [Anger-Hostility]	48.5 ± 11.1	44.7 ± 8.5	0.003
	CB [Confusion-Bewilderment]	51.5 ± 7.6	46.8 ± 7.7	0.008
	DD [Depression-Depression]	51.8 ± 10.2	48.6 ± 9.2	0.003
	FI [Fatigue-Lethargy]	52.3 ± 11.7	45.8 ± 8.2	0.006
	TA [Tension-Anxiety]	49.0 ± 9.0	45.4 ± 7.5	0.012
	VA [Vitality-Vitality]	48.3 ± 9.8	50.9 ± 10.9	0.113
	F [Friendship]	48.0 ± 8.6	51.2 ± 9.8	0.083
	TMD score	51.1 ± 9.6	46.2 ± 7.9	0.003
STAI	State anxiety/P scale	44.4 ± 9.4	43.3 ± 8.0	0.789
	State anxiety/A scale	49.7 ± 12.4	48.6 ± 8.8	0.504
	State anxiety total scale	44.3 ± 10.3	43.1 ± 7.1	0.455
	Trait Anxiety/P Scale	45.5 ± 10.5	40.3 ± 10.9	0.005
	Trait Anxiety/A Scale	53.4 ± 11.0	50.8 ± 10.7	0.074
	Trait anxiety total scale	49.2 ± 10.7	44.9 ± 9.4	0.008
	5 stages/state anxiety/P scale	2.3 ± 1.1	2.3 ± 0.9	1.000
	5 stages/state anxiety/A scale	3.1 ± 1.2	2.8 ± 0.8	0.180
	5 stages/state anxiety/full scale	2.8 ± 0.9	2.3 ± 0.7	0.059
	5 stages/trait anxiety/P scale	2.4 ± 1.1	2.1 ± 1.0	0.103
	5 stages/trait anxiety/A scale	3.4 ± 1.2	3.0 ± 1.2	0.059
	5 levels	3.0 ± 1.0	2.6 ± 1.0	0.025

Results are expressed as mean ± SD, n = 12. OSA, obstructive sleep apnea (OSA) sleep inventory MA version; POMS2®, Profile of Mood States 2nd Edition; Japanese version; STAI, State-Trait Anxiety Inventory-Form JYZ; SD, standard deviation.

(p = 0.003,) factor 2 (initiation and maintenance of sleep) (p = 0.004,) factor 4 (refreshing feeling) (p = 0.003,) and factor 5 (sleep duration) (p = 0.034.)

Assessments in subjects' feelings using POMS2® were perfomed using seven scales: "Anger-Hostility," "Confusion-Bewilderment," "Depression-Dejection," "Fatigue-Inertia," "Tension-Anxiety," "Vigor-Activity," and "Friendship". Additionally, "TMD scores" were used to represent negative feelings in an integrated manner. As the first step of the assessment, raw scores from the examinations were converted into T scores. This was done for the standardization of measurements, with a mean value of 50 and a standard deviation of 10. Next, a statistical analysis was performed on the T scores of each scale. As a result, the following items showed significant decreases in assessments of test product usage compared to non-usage: AH: Anger-Hostility (T score, p = 0.003), CB: Confusion-Bewilderment (T score, p = 0.008), DD: Depression-Dejection (T score, p = 0.003), FI: Fatigue-Inertia (T score, p = 0.006), TA: Tension-Anxiety (T score, p = 0.012) and TMD score (T score, p = 0.003). Additionally, a trend toward an increase in T score (p = 0.083) was observed for F: Friendship during the test product-usage period compared to the non-product-usage period.

The STAI confirmed significant decreases during the test-product-usage period in the T scores for the "trait anxiety P scale" (p = 0.005) and the "trait anxiety total scale" (p = 0.008), as well as in the percentiles for the "trait anxiety P scale" (p = 0.005) and the "trait anxiety total scale" (p = 0.008). In addition, significant decreases were observed in the five-level rating (T score) of the "trait anxiety total scale" (p = 0.025) and in the five-level rating (percentile) of "trait anxiety total scale" (p = 0.008).

Biomarker assessment

Salivary oxytocin (OT)

Salivary OT levels at bedtime and wake-up time did not show significant differences during the test-product-usage period compared to the non-test-product-usage period (***Table 2***). The increase rate of salivary OT at bedtime indicated a positive trend, however, significant differences were not observed (p < 0.1).

Data analysis results by sex are shown in ***Table 3*** and ***Fig. 1***. There was no significant difference in male subjects. Salivary OT levels in female subjects during the test product usage period significantly decreased at wake-up time compared to the non-test product usage (2.21 ± 0.25 pg/mL 1.83 ± 0.39 pg/mL, p = 0.028). One week after the commencement of test product usage, salivary OT levels were significantly higher at bedtime compared to wake-up time (1.83 ± 0.39 pg/mL vs. 4.65 ± 2.11 pg/mL, p = 0.035).

Urinary oxytocin (OT)

A high trend in urinary OT was observed when comparing test mattress usage and non-usage (p = 0.065, ***Table 2***), however, no significant differences were observed.

Table 2. Measurement results of OT, melatonim and catecholamine metabolites.

Item		Unit	Before Mean ± SD	After 1 week Mean ± SD	p value
Salivary OT	(Bedtime)	pg/mL	3.81 ± 2.85	4.78 ± 2.62	0.324
	(Wake up time)	pg/mL	2.44 ± 0.93	2.83 ± 2.15	0.525
Urinary OT		pg/mL	18.81 ± 7.96	20.66 ± 7.19	0.208
Urinary melatonin		ng/mL	23.91 ± 18.40	25.36 ± 16.71	0.727
Urinary catecholamine metabolites	Dopamine	mg/gCr	0.22 ± 0.06	0.23 ± 0.07	0.640
	Serotonin	mg/gCr	0.10 ± 0.02	0.10 ± 0.02	0.794
	GABA	mg/gCr	0.25 ± 0.08	0.28 ± 0.19	0.455
	5-Hydroxyindoleacetic acid	mg/gCr	3.29 ± 0.75	3.47 ± 1.14	0.508
	HVA	mg/gCr	4.23 ± 1.19	4.68 ± 1.25	0.077
	VMA	mg/gCr	3.20 ± 1.07	3.45 ± 1.21	0.042

Results are expressed as mean ± SD, n = 12. OT, oxytocin; GABA, γ-aminobutyric acid; HVA, homovanillic acid; VMA, vanillylmandelic acid; SD, standard deviation.

Table 3. Salivary OT: Analysis by gender.

Item		Unit	Before Mean ± SD	After 1 week Mean ± SD	p value
Salivary OT	(Bedtime)				
	Female	pg/mL	3.18 ± 1.46	4.65 ± 2.11	0.138
	Male	pg/mL	4.44 ± 3.45	4.92 ± 2.85	0.798
	(Wake up time)				
	Female	pg/mL	2.21 ± 0.60	1.83 ± 0.39	0.028
	Male	pg/mL	2.66 ± 1.06	3.83 ± 2.52	0.356

Results are expressed as mean ± SD, female n = 6, male n = 6. OT, oxytocin; SD, standard deviation.

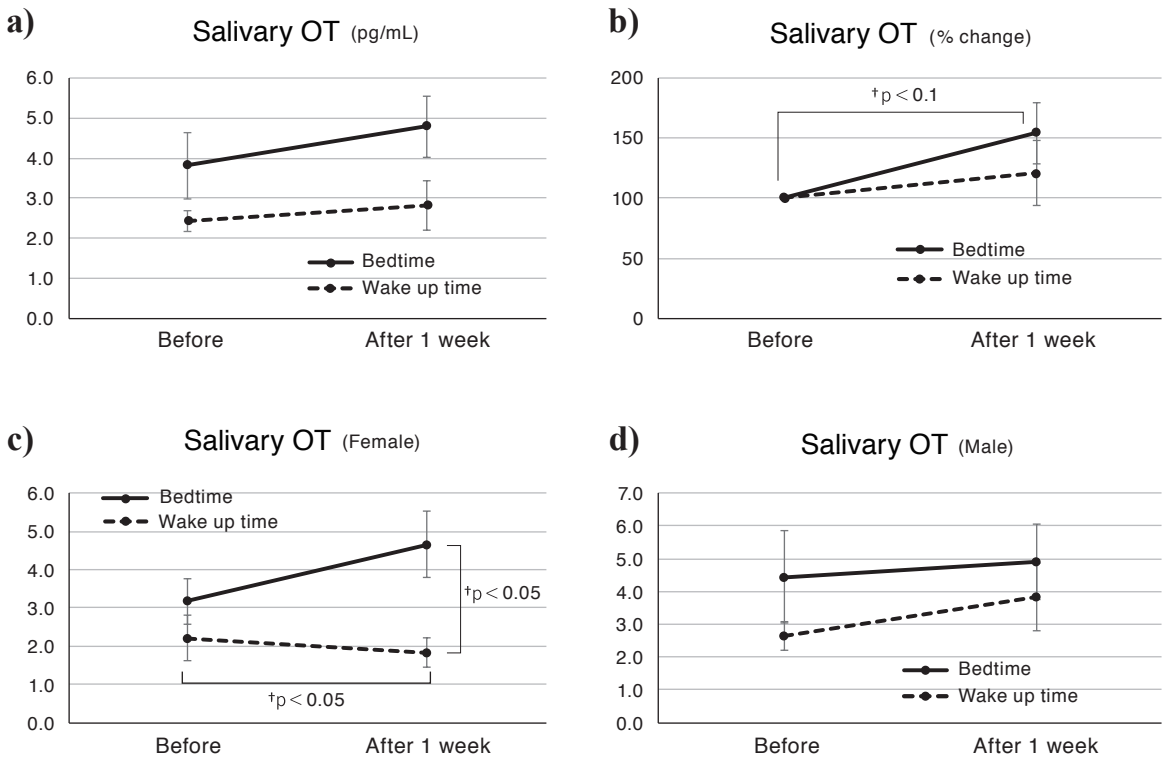

Fig. 1. **Trend graph of salivary OT: Bedtime and wake-up time.**

a) Measurement values: the total of males and females (n = 12). **b)** Rate of change: the total of males and females (n = 12). **c)** Measurement values: females (n = 6). **d)** Measurement values: males (n = 6). Results are expressed as mean ± SEM. OT, oxytocin; SEM, standard error mean.

Table 4. Urinary HVA and VNA: Analysis by gender.

Item		Unit	Before Mean	Before SD	After 1 week Mean	After 1 week SD	p value
Urinary catecholamine metabolites	HVA						
	Female	mg/gCr	5.03	± 0.77	5.60	± 0.80	0.115
	Male	mg/gCr	3.43	± 0.86	3.77	± 0.75	0.182
	VMA						
	Female	mg/gCr	3.69	± 1.15	4.15	± 1.25	0.035
	Male	mg/gCr	2.71	± 0.52	2.74	± 0.37	0.698

Results are expressed as mean ± SD, female n = 6, male n = 6. HVA, homovanillic acid; VMA, vanillylmandelic acid; SD, standard deviation.

Urinary melatonin

The level of urinary melatonin did not show a significant difference between non-usage (23.9 ± 5.3 ng/mL) and usage of test mattresses (25.4 ± 4.8 ng/mL). The rate of change was shown in ***Table 2*** (p = 0.155).

Urinary stress markers, six components analysis (dopamine, serotonin, GABA, 5-hydroxyindoleacetic acid, homovanillic acid, and vanillylmandelic acid)

Vanillylmandelic acid (VMA) significantly increased during the test-mattress-usage period (p = 0.035).

Homovanillic acid (HVA) had a trend toward an increase during the test mattress usage period (p = 0.077).

Discussion

Improvement effects on "Sleep quality": Comparison among previous studies

We have conducted clinical trials for this test product six times in total, and the present trial is the seventh study [5-10].

For PSQI-J in the six previous studies, sleep quality significantly improved in all six studies. Sleep latency significantly improved in five out of six, sleep duration and daytime dysfunction in four out of six, and sleep disturbance in three out of six studies. The global score for the total assessment in PSQI-J (PSQIG) significantly improved in all six studies.

The OSA sleep inventory conducted in the first, third, fourth, and sixth studies showed significant improvements

of factor 1and factor 4 in all four studies, of factor 2 in three out of four, and factor five in once out of four studies.

The previous studies have confirmed similar results and improvement effects on subjective symptoms is highly reproducible. It has been suggested that the usage of this test product, the mattress, improves sleep quality. However, the sixth study had only female subjects who had subjective symptoms of menopausal syndrome so that their mental and physical stresses before bedtime, which could be related to menopause, would be severe. Therefore, items of improvement effects in the sixth study were slightly small in number [10].

Overviewing transitions of subjective symptoms in the present study, significant improvements were confirmed in a large number of items due to the usage of appropriate bedding. The data results from the OSA sleep inventory indicated improvements in sleep quality. Findings of POMS2® suggested that factors of anger, depression, fatigue, anxiety decreased, and subjects felt better. It is considered that suppressed mental and physical stresses were reduced. The STAI is, in particular, an evaluation scale, which focuses on "anxiety" as strongly related to mental and physical stresses. This scale is characterized by divided items: state anxiety (temporary reactions to a situation which causes anxiety) and trait anxiety (tendency for relatively stable reactions toward experiencing anxiety) as well as anxiety-present items, P-items, and anxiety-absent items, A-items. The present study confirmed that trait anxiety, as measured by the center of the P-scale, significantly improved. The subject group of the present study had a clear anxiety factor related to deterioration in sleep quality due to "defects in bedding". Mental and physical stresses, which was related to the deterioration of "sleep quality", had been continuously imposed on the subjects (*Fig. 2*). However, it was unknown how long stress burden had been imposed.

In analysis, it is important to accurately evaluated what stage of the stress respond process the attribute of subject group was placed in.

Regarding Oxytocin (OT)

OT, which is a peptide consisting of nine amino acids (Cys-Tyr-Ile-Gln-Asn-Cys-Pro-Leu-Gly), is produced in neurons in the hypothalamus of the diencephalon. It is secreted into the bloodstream as a hormone from the nerve terminal which stretches to the neurohypophysis, and acts on the peripheral nerve [11]. Furthermore, OT functions in the central nerve as a nerve transmitter substance and neuroregulatory. Blood OT, via the Receptor for AGEs (RAGE) on capillary endothelial cells of the blood-brain barrier (BBB), moves into the brain and reinforces the central nervous system effect [1,2,12,13].

The hormone`s primary function is to induce uterine contractions during childbirth and the milk ejection reflex during breastfeeding. Synchronous bursting emerges in a

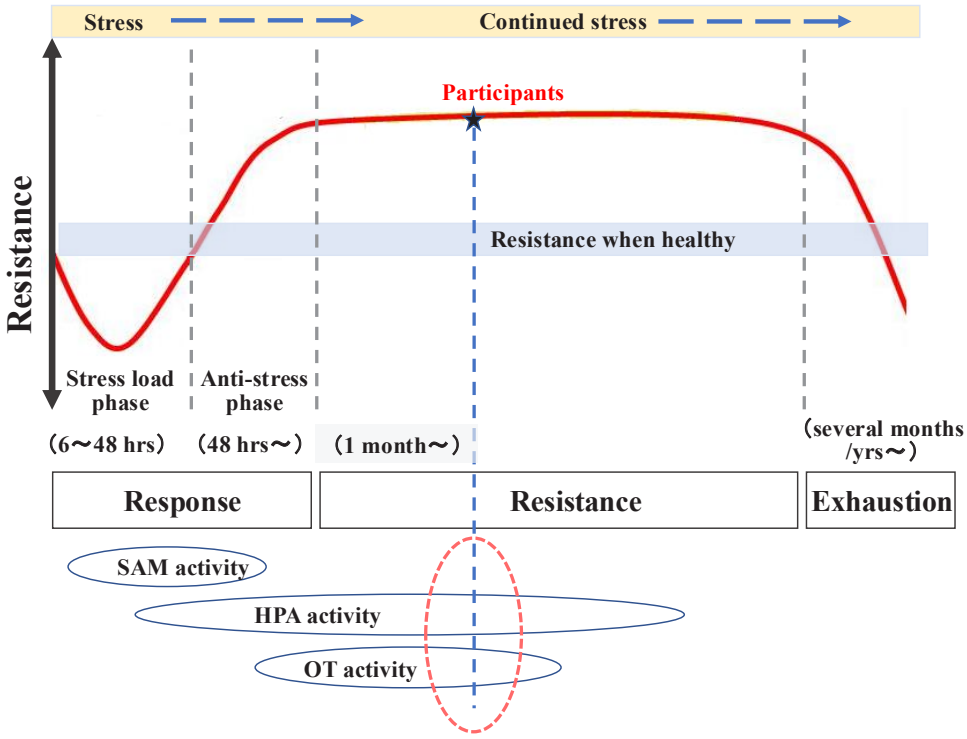

Fig. 2. **Temporal change after stress imposition.**

Before interventions were provided to the subjects, subjects were placed in the resistance period, where OT secretion was accelerated in response to stresses related to "sleep quality", SAM activity was subdued, and the parasympathetic nerve was dominant (in the area circled with a red dotted line). SAM, sympathetic nervous adrenal medullary system; HPA, hypothalamic-pituitary-adrenal axis; OT, oxytocin; OT activity, OT neuron activity.

neuronal network in the hypothalamus. A large quantity of OT secretes into the bloodstream from the axon terminal projecting to the neurohypophysis[14]. To express love during child rearing in the breastfeeding period, it is necessary for blood OT to be transferred into the brain through the BBB via a transporter. It has been confirmed that RAGE on capillary endothelial cells, which make up the BBB, plays a significant role as an OT transporter[12].

OT is independently produced in the cell body of the OT neuron (a small cell neuroendocrine neuron of the parvocellular neurosecretory cells). These neurons are designated as autonomic neurons; some of these neurons project axonal fibers to preganglionic sympathetic neurons in the brain stem and the spinal cord. OT is related to the control of autonomic nervous functions. These neurons are related to the adjustment of the autonomic nervous system[15]. Central nervous system effects are integrated effects of brain-derived and blood-derived OT.

The central action of OT is related to higher brain functions, such as bond between parent and child, and feelings of trust and confidence. It enhances comfort and peace fostering both mental and physical bonds, which has been drawing attention recently[16-18]. OT enhances the alleviation of anxiety, sedative effects, and the reduction of mental and physical stresses. Furthermore, an increased sense of confidence and empathy strengthens the bond. Human relationships influence life satisfaction. There is a significant positive correlation with happiness and social relations. OT may serve as a mechanism that promotes social relationships, thereby fostering happiness[19,20].

OT secretion is known for a stimulating factor. Stimuli that stimulate OT secretion in mothers include tactile stimuli on mammilla from breastfeeding, visual stimuli from making eye contact with a baby, olfactory stimuli from the scent of a baby, and auditory stimuli from the cry of a baby. Blood OT concentration levels are higher in breastfeeding than formula feeding. Mild skin stimulation from massages[22] and stimulation from physical exercise affect OT secretion. Furthermore, OT is affected by sex hormones, and OT secretion is increased by estrogen administration to women and testosterone administration to men[23]. However, human emotions are complicated and the OT secretion system is not simple. Touch between partners mitigates negative feelings, sometimes evokes pleasant sensations, and serves as a reward, which promotes OT secretion and further expectations[24]. OT is secreted in anticipation. However, touch with an unfavorable person leads to feelings of aversion. When hope is betrayed, OT is not secreted. Instead, OT reacts to clear stress stimuli such as harmful stimuli, stimuli with fear-inducing conditions, and exposure to new circumstances, which induce OT secretion from the pituitary gland[25].

We humans, including the subjects in the present study, are active during the daytime and receive stimuli such as favorable stimuli and stressors. Therefore, salivary OT levels are higher at bedtime than upon waking under physiological conditions. However, it is considered that under conditions with stress imposition, "response resistance" is exacerbated and stimuli (both favorable stimuli and stressors) are unlikely to induce OT secretion. It is recognized that mothers with intense stress tend not to feel love toward babies. OT secretion induced by stimuli from breastfeeding, would be minimal.

The finding that female subjects showed a significant decrease in OT values upon waking after the interventions would suggest that baseline values also had some important meanings. One possibility is that baseline levels could rise in a case where stimuli of unfavorable stress chronically continued. Another possibility is that high quality sleep reduces salivary OT levels, which had been raised by diverse stimuli, thereby lowering the baseline. In a condition of lowered baseline in this manner, "response resistance" is mitigated, so that OT secretions can easily occur for favorable stimuli and stressors.

The results of this trial confirmed that salivary OT levels of females, measured at the pre-activity baseline, significantly decreased by "improving the quality of sleep." It is possible that under conditions of decreased baseline, similar to the condition in the present study, the responsive resistance of OT secretion is alleviated allowing for diverse activities during the daytime and related favorable stimuli to promote an increase in volume of responsive OT secretion.

The meanings of baseline are discussed in a following section in this study, "Comparison in salivary OT levels between at bedtime and wake-up time."

Circadian rhythm an Oxytocin (OT)

Adaptations of living bodies to their environments are controlled by circadian clock network. This network is related to multiple hormones. The sleep/wake cycle is strictly controlled by melatonin, orexin, and cortisol. Food ingestion is influenced by the circadian adjustment of leptin, ghrelin, insulin, and orexin. The discrepancy between circadian rhythms and the Earth's rotation is adjusted by energy intake (such as the rise of blood glucose levels in the morning and elevation of blood amino acids), exposure to morning sunlight, and suppression of melatonin formation. Maintaining sleep quality is highly important for the homeostasis of secretions of these hormones.

Melatonin is produced by the pineal gland during night. Blood melatonin levels alter in a 24-hour cycle. Melatonin, which is associated with preparing for sleep, halts secretions due to exposure to bright sunlight in the morning, maintains low levels in the morning, and rises from the evening to bedtime (from 21:00 to 22:00)[26-28]. Various studies have reported that melatonin at night is associated with sleep quality and overall happiness. Furthermore, melatonin is linked to depression and some antidepressants raise blood melatonin levels[29].

It is recognized that blood OT is not highly affected by circadian rhythms. However, it is necessary to consider the measurement time and menstrual cycle. While blood OT levels of men are low at bedtime compared to wake-up time, blood OT levels of women during the incubation period tend to be low at wake-up time similar to men, but in some cases during the incubation period, sudden spikes in OT levels are observed at wake-up time[23].

Mental and physiological stresses and Oxytocin (OT)

The stress response involves psychological processes that react to imbalances due to stressors of extraneous stimuli, progressing through the stress load phase, anti-stress phase, resistance, exhaustion, and recovery. Human bodies show non-

specific responses regardless of types of stressors. Responses to stressors are different, from one individual to another, depending on age, sex, and past experiences. Information of mental and physical stresses are transmitted, via the cerebral cortex and the limbic system, to the hypothalamic area in the diencephalon. Through two systems' paths, the sympathetic nervous adrenal medullary system (SAM) and the hypothalamic-pituitary-adrenal axis (HPA axis), stress response symptoms are induced. Followed by this reaction, neuroendocrine control and immunomodulation are implemented to aid in recovery and restoration, alleviating symptoms[25]. Responses of the human body to mental and physiological stresses consists of two reactions: stress load phase (responses that directly disrupt homeostasis) and anti-stress phase (recovery responses that correspond to these stresses (*Fig. 3*).

OT is produced in the soma of OT neurons in the hypothalamus, transferred through the axon, and released into the bloodstream from the posterior pituitary in response to physical and mental stress stimuli[25]. OT is secreted in response to various stimuli such as emotional changes in the human body. OT plays a role in alleviating responses mainly toward anxieties and stresses. Furthermore, OT is secreted in response to disturbances in homeostasis that are caused by mental and physical stress, and promotes resistance to stresses, and serves to maintain homeostasis. Therefore, OT is regarded as a significant hormone.

Similarly, vasopressin, which is a posterior pituitary hormone, is secreted from the posterior pituitary, is secreted in response to physical stress stimuli such as acute pains[30] and vigorous exercise[31]. Vasopressin has different behaviors from OT and is rather suppressed by mental stress stimuli inhibiting its secretion[30]. It has been reported that rodents have differences in vasopressin between the two sexes, and male rodents increase aggressiveness by vasopressin[32]. From a social perspective, this is understandable to protect their herd, a collective community. It has been reported that individuals with a mutation in the vasopressin-receptor-related gene, which is associated with pair-bonding behavior in humans, have difficulties in their marital life, and supposedly have a higher rate of non-marriage[33].

Blood OT is a marker for vasopressin activity in the hypothalamus and/or OT activity. OT is secreted not only from the posterior pituitary gland into the peripheral blood, but also from the soma into the hypothalamus[34]. Blood OT, via RAGE, transfers across the capillary endothelial cells of the BBB from the bloodstream into the brain. Blood-derived OT is added to OT released in the hypothalamus, and synergy is promoted to address homeostasis disorders due to mental stress.

During this trial, improvements in "sleep quality" did not alter salivary OT levels in men. However, salivary OT levels in women significantly decreased at wake-up time (*Fig. 1*). The subjects of the present study consisted of individuals who had experienced mental stress due to bedding defects and were in a condition where OT response secretion was stimulated to maintain homeostasis. The usage of appropriate bedding improved "sleep quality" and stresses were reduced in this study. As a result, it was considered that OT secretion response was reduced and the secretion quantity was lowered.

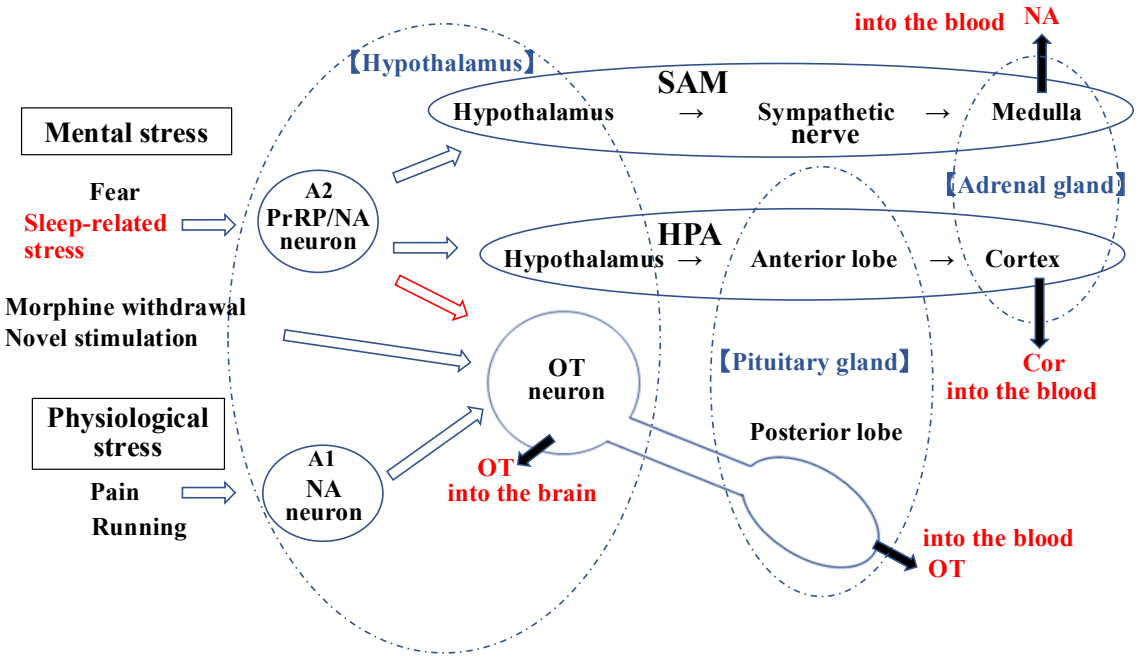

Fig. 3. **Transmission pathway of stress response.**

Stress is classified into two groups, psychological and physiological stress. Both types of stress stimulate OT neurons, promote the response OT secretion, and respond to homeostasis disruption. Mental stresses are classified into prolactin-releasing peptide/noradrenaline, PrRP/NA neuron dependence and independence. The former influences the whole body via the SAM and HPA. Stress that reduces "sleep quality" would belong to the former. This is addressed with Reference 20, 25, 28, and 30. SAM, sympathetic nervous adrenal medullary system; HPA, hypothalamic-pituitary-adrenal axis; OT, oxytocin; NA, noradrenaline; Cor, cortisol; PrRP, prolactin-releasing peptide.

Stress and behaviors of catecholamine

Stress responses and the two pathways of the SAM system and HPA axis have been mentioned above. Activation of the SAM system induces the release of catechalomines (adrenaline: AD and norepinephrine: NA) into the blood. Elementary reactions are induced such as blood pressure elevation, perspiration, increase in blood glucose levels, an alert state, and combat readiness. The brainstem (midbrain, pons, medulla oblongata, and diencephalon) have many neurons; A6NA neurons in the locus coeruleus of the pons, A1NA neurons in the ventrolateral part of the medulla oblongata, and A2 PrRP/NA neurons in the dorsomedial region of the medulla oblongata. These neurons are activated by diversified stresses [25, 35]. Norepinephrine (NA) and Prolactin-Releasing Peptide (PrRP) coexist in the PrRP/NA neuron. Activation of the HPA axis induces the release of glucocorticoids such as cortisol into blood. Diversified functions are influenced such as blood pressure elevation, an increase in blood glucose levels (increases in gluconeogenesis), increasing cardiac contractility, increasing cardiac output, and immune system regulation (inflammation suppression). Cortisol receptors are in the hypothalamus, the hippocampus, and the pituitary gland. When the amount of cortisol secretion increases, excessive stress stimuli are not loaded via the negative control system [35].

When acute stresses persist, the responsive reaction enters the resistance period. The balance between continuous stressors and the capacity for resistance is maintained and the biological defense reactions are established (***Fig. 2***). The conditions of the subjects before the interventions of this trial were assumed to be as follows: the homeostasis of the subjects was maintained via the activation of resistance, as mild stresses were continuously imposed on the subjects. Subjects had not been in the stage of exhaustion yet. There is a time lag between the activations of the SAM system and HPA axis. Sympathetic nervous adrenal medullary system, SAM, precedes HPA, and SAM has a shorter duration than HPA. Therefore, in the second half of the response period, SAM activities decrease, and cortisol secretion becomes dominant via the activation of the HPA.

When the stress condition continues further, the power of resistance gradually declines and homeostasis breaks down. Therefore, stress adjustment disorders occur in the period of exhaustion. When the secretion of cortisol continues due to excessive stress, the hippocampus and glial cells are damaged. It has been reported that clinical depression has symptoms such as hippocampal volume loss and decreased neurogenesis [36].

Vanillylmandelic acid (VMA) and homovanillic acid (HVA), which are catecholamine metabolites, are detected in the urine. Measurement of VMA is used to diagnose neuroblastoma in the pediatric period and melanocytoma in adolescence. Blood VMA levels fluctuate with circadian variation, showing high levels during the day and low levels at night. Catecholamine secretion is increased by factors such as body position, exercise, hypoglycemic stimulation, and stress. Consequently, VMA is affected by these factors.

Previous studies with animal experiments have reported that OT neurons would activate the sympathetic nerve in the following manner: OT neurons project to the supramammillary nuclei, activating recognition functions [37], and act on the rostral medullary raphe region (rMR) activating the sympathetic nerve [38]. Rostral medullary raphe regions act on brown cells, elevating body temperature, increasing heart rate, increasing basal metabolic rate, and suppressing obesity [38]. Obesity is related to leptin. In mice with leptin tolerance, OT suppresses hyperphagia and improves obesity [39]. In humans, mental and physiological stresses generally induce obesity due to overeating [39-42]. It is assumed that in the early state of stress impositions, the SAM pathway is activated. However, over time, HPA is activated, OT neurons are activated, and then, the activated SAM pathway gradually subsides. Consequently, the basal metabolism rate decreases, which leads to obesity.

The present trial confirmed a significant increase of VMA and a trend of increasing HVA. It was considered that subjects were in a balanced state between continuous stresses due to bedding defects and resistance of the body, where biological defense reactions are balanced and the autonomic nerve system is in equilibrium. The formation of AD and NA, which are biomarkers for the autonomic nerve activation, is inhibited and consequently, VMA and HVA showed low levels. Interventions in this trial reduced stressors due to "inappropriate bedding" and biological defense reactions were alleviated. Therefore, the autonomic nerve activity, which had been suppressed, regained function. As a result, this led to increases in VMA and HIV.

Response pathway of "sleep quality"-related stress

As mentioned above, stresses are classified into two groups, psychological and physiological stress. Both types of stresses stimulate OT neurons, promote the response OT secretion, and respond to disturbances in homeostasis. (***Fig. 2***) [25, 30, 35, 43]. Mental stresses are classified into prolactin-releasing peptide/ noradrenaline, PrRP/NA neuron dependence and independence. The former influences the whole body via the SAM and HPA axis. It had been unclear which types of stress stimuli were related to the deterioration of "sleep quality". Results of this trial confirmed that the improvement in "sleep quality" significantly changed both NA-derived catecholamine metabolism and OT secretion. The present study suggested that stresses related to the deterioration of "sleep quality" would be PrRP/NA neuron dependence.

Previous studies have suggested a hypothesis that biological stress responses are broadly divided into two categories, depending on the types of reactions: a loading pathway, where the impacts of stress stimulation pathway are transmitted throughout the whole body, and a recovery pathway, where the disruption of homeostasis is addressed [25]. Findings of the present study could support this hypothesis. The present study has confirmed that via the reduction of stresses which are related to lowered "sleep quality", salivary OT levels, which was a biomarker, were decreased. Thus, the index for the accelerated anti-stress pathway was decreased. In addition, urinary VNA levels were decreased, which meant the increase of NA, where the suppressed SAM activity of the sympathetic nervous system was recovered.

As previously reported in our studies, it was observed in similar trials that an increase of NA and decrease of serotonin were induced through the mitigation of stresses of deteriorated "quality of sleep".[5] We had difficulties interpreting these phenomena at that time. We now

understand that when the stressors of "inappropriate bedding" were reduced, biodefence reactions were alleviated, suppressed sympathetic nerve activities regained their function, and consequently, NA levels in the blood increased. Serotonin, similar to OT, plays a role in stress resistance. It is interpreted that serotonin levels were decreased as an aspect of the activities in the recovery pathway.

Furthermore, similar trials have observed that the mitigation of stresses related to deteriorated "quality of sleep" induced a decrease in blood cortisol levels and a decrease in dehydroepiandrosterone sulfate (DHEA-s)[6]. The former was induced by the reduced activity of the stimulation pathway (HPA) due to stress alleviation. DHEA is known as an anti-stress hormone[44, 45]. Similar to OT and serotonin, DHEA, which has stress resistant effects, decreased as an aspect of the reduced activities in the recovery pathway.

Comparison in salivary oxytocin (OT) levels between at bedtime and wake-up time

Alterations in salivary OT levels between bedtime and wake-up time are shown in ***Fig.1***. As a premise, it was postulated that stresses associated with a decline in "sleep quality" were alleviated after using the mattress. Items which showed significant differences ($p < 0.05$) were focused on, and trends ($p < 0.1$) were considered. Other items were regarded as no change.

Comparisons between bedtime and wake-up time showed that salivary OT levels seemed to be lower at wake-up time. However, only women`s OT levels after the interventions were significantly lower. In the investigation after the interventions, salivary OT levels were first tested at bedtime, and another test of salivary OT levels was later conducted at wake-up time after sleep. We proceed the discussion under the premise that in the reverse order, the values for bedtime are the same. Every activity and every action during daytime are stimuli toward the body. Pleasant stimuli (e.g. tactile stimuli, visual stimuli, and olfactory stimuli in comforting a baby) increase OT secretion. In addition, uncomfortable stimuli, as a stress response, increase OT as compensation. The reason for high-levels of salivary OT at bedtime is that various stimuli from daytime activities have accumulated. It is considered that the levels return to the baseline during sleep without stimuli and stresses. Therefore, the level of salivary OT at wake-up time is assumed to be equivalent to the baseline. It can be interpreted as follows: In a case where OT levels are largely increased because of uncomfortable stresses, "healing effects due to sleep" are at work. Contrarily, in a case where OT levels are increased because of stimuli based on affection, "reset due to sleep" occurs. A state of decreased baseline would indicate a condition in which "responsive resistance" is alleviated and secretion capacity of OT is recovered due to stimuli.

Taking women as an example, OT levels at wake-up time significantly decreased via the interventions, and OT levels at bedtime were significantly higher than those at wake-up time. OT levels at bedtime did not show changes in women but an increasing trend was shown in the total of men and women. It is reported that a half value period in the blood of goats is approximately 20 minutes[46] while the half value period of salivary OT in humans is 30～40 minutes[47]. Therefore, OT secretion stimuli, which affects measurement values at bedtime, derive from an event close to bedtime. In particular, "feelings of hope for an upcoming event" would be a large factor of favorable stimuli for OT secretion. The present study did not conduct an in-depth interview survey. It is assumed that subjects in the present study had the common factor of "feelings of hope for sleeping with comfortable bedding", as there might have been a possibility of a romantic factor.

The mechanism behind the decrease in OT levels at wake-up time has been mentioned above. The increasing trend in the total group of men and women is assumed to be induced by the increase of OT volume as follows: chronic stress and the deterioration of "sleep quality" were alleviated by the usage of comfortable bedding, the "responsive resistance" of OT secretion was reduced, and the OT which was secreted by various stimuli during daytime increased in quantity.

Issues of salivary examination

One method for assessing mental and physiological stresses is to use a salivary biomarker[48]. This method is non-invasive and simple to measure. Therefore, a high expectation is held in clinical settings. However, there are points to note regarding accuracy. In comparison with blood biomarkers, accuracy and reproducibility should be reconsidered. Saliva is secreted from the major salivary glands such as the parotid gland, submaxillary gland, and sublingual gland, as well as from various minor salivary glands. Saliva, which is a mixture of serum and mucus, is controlled by both the sympathetic nervous system and the parasympathetic nerve system. Saliva secretion volume, viscosity, moisture, and concentration fluctuate with considerable individual variation[35]. These points are distinctly different from blood components, which maintain homeostasis. For example, using mouthwash before saliva collection tends to lower the concentration due to attenuation. The viscosities of saliva are distinctly different between the state of slight dehydration following perspiration and the state of slight swelling immediately after waking up. Calibration methods employing a correction factor have not been established, although creatinine correction is employed for urine samples.

Diurnal variation in salivary cortisol concentration of the same individual follows similar diurnal variation in blood concentration. Examinations of four subjects in diurnal variations showed similar circadian variations. However, comparisons in salivary concentrations among individual subjects showed large differences. Values of salivary cortisol concentration vary in four to five distinct amounts. Therefore, measuring salivary samples is required to set a normal range standard for each individual. The assessment method using standardized values cannot be applied to salivary samples; the concentration range standard, which is used in blood examinations, is not appropriate[35]. In cortisol measurement, other than diurnal variation, lifestyle factors, such as exercise, diet, excretion, bathing, work, and mental and physiological stresses influence the SAM and HPA. Consequently, the attributes of saliva change independently of cortisol secretion. In comparison to using blood biomarkers, stress evaluations employing salivary biomarkers could be difficult to assess at a specific point in time. However, when assessing changes

over time and changes before and after interventions of the same individual, a salivary biomarker can be effective to some extent.

Therefore, to provide a specific assessment at a specific point, using salivary OT concentration measurement as an index is difficult to employ. However, it would be effective to measure increase/decrease or fluctuation of OT before and after intervention.

Multiple studies have reported on blood OT concentration. Fasting blood plasma OT concentrations in women largely fluctuate depending on a gestational period or an estrus cycle[23,48]. Fluctuations during an estrus cycle are significantly lower during the luteal phase (2.1 ± 1.3 pg/mL) than during the follicular phase (4.5 ± 2.6 pg/mL, mean ± SEM) (n – 22). Men's fasting blood plasma OT concentrations are significantly lower (3.5 ± 1.7 pg/mL, n = 51) than that of women in the luteal phase[23]. It is reported that regarding diurnal variation of both men and women, concentration levels are low with a slightly low trend during the awakening period compared to during sleep.

Some studies have also reported on comparisons between blood and salivary OT. The mean blood OT concentration was 471.8 ± 314.4 pg/mL and the mean salivary OT concentration was 61.3 ± 36.0 pg/mL. Its correlation was r = 0.485 and p = 0.026[49]. The coefficient of variation for the measurement was within 10 %.

The present trial employed the evaluation index. The following are the results of the coefficient of variation. (CV = the standard deviation/the mean):

Salivary OT (at bedtime)	0.75
Salivary OT (at wake-up time)	0.38
Salivary OT (bedtime – wake-up time)	1.81
Salivary OT (bedtime + wake-up time)	0.65

Compared to four- to five-fold variance in salivary cortisol concentration, variances in values of salivary OT concentrations seem to be minor. Under adjusted conditions at wake-up time, CV was slightly low at bedtime. Still, the value was larger (0.38 or larger). As reasons for the decreased levels of salivary OT have been explained in the discussion section, it is assumed that improvements in stresses related to "sleep quality decline" induced a decrease in OT responsive secretion. Another possibility, where female subjects had less tactile stimulation (after the bedtime assessment), was unable to be clarified.

Differences in salivary OT between men and women

The difference in salivary OT between men and women has been reported as follows: men (n = 43) 594.8 ± 123.3 pg/mL (mean ± SEM), median value 235.4 pg/mL, and women (n = 15) 1444.7 ± 693.0 pg/mL, median value 430.3 pg/mL[50]. The measurement values of women had large individual differences (SEM and CV). Methods for salivary sample collections were not established regarding fluctuations due to the estrous cycle and the circadian rhythm based on hormone dependency. The present study showed that the CVs for women were 0.46 and for men were 0.78 (salivary samples at bedtime), 0.27 for women and 0.40 for men (salivary samples at wake-up time), and 0.21 for women and 0.66 for men (urinary samples). The present study had minor variations in the data for female subjects, as they were limited to postmenopausal women. Therefore, fluctuations due to the menstrual cycle were avoided.

The effects of improvements in stresses related to "sleep quality deterioration" were shown in ***Table 5***. Differences between men and women were confirmed in the results of the STAI and POMS2. In women, there were six items showing significant improvements. However, men had only one item, POM2 "Fatigue-Inertia". Women had more items of improvement. The reason was unclear. It was certain that there was a minor individual difference in women. According to a study on FACIT-Fatigue values by sex and age, higher-level fatigue was observed in women than men for all age groups (except for the age group of 65 ~ 74)[51]. From this report, it has been suggested that women are more susceptible to stresses with fatigue in general. Thus, the present study suggested that women would be more likely to show improvements in the data results.

In animal experimental research, it was reported that two subtypes of OXTR neurons causally control consolation or aggression that may underlie the switch between consolation and aggression, for example soothing stressed companions and being aggressive to intruders[52]. These activations of neurons depend on sex and are related to character developments through communal living for mice[53].

There is also another report on OT. OT serves to strengthen bondages among companions on one side, and to be aggressive to opponents on the other side[54]. Research has been conducted on differences of behavioral expressions such as aggressiveness between men and women. This is related to the actions and interactions of sex hormones such as estrogen and androgen, as well as neuroendocrines such as OT, vasopressin, and serotonin[55]. It is commonly stated that men tend to be more aggressive than women. This would be attributed to the influence of androgen. However, it is not so simple. Intranasal administration of OT induces, regardless of sex, temporary elevation of aggressiveness, as a study has reported[56,57]. In other studies on intranasal administration of OT[58,59], a reduction in aggressiveness has been distinctively observed in men.

Child abuses, such as neglect, increase the risk of developing emotional disturbances later in life. It has been reported that abuses are related to mutations in the OT receptor gene, in particular, a decrease in methylation[60]. When emotional expressions are evaluated regarding OT, it is necessary to consider the expression and mutation of OT receptors.

Future Prospects

OT is used as a drug for labor induction. Labor induction is the use of medications to start and progress labor in a pregnant woman whose labor pains have not started. This medication is administered to a pregnant woman with deficient OT secretion when the due date has passed or after amniorrhexis. The reasons for deficient OT secretion at the time of delivery in mothers are not clear. Aging and menopause[61], excessive weight gain during pregnancy[62], mental and physiological stresses[63], and malnutrition are considered to be reasons. The present study suggests possibilities that chronic stress may lead to the reduction of OT secretion activities. We expect that the mitigation of mental and physiological stresses due to improvements in "sleep quality" would positively affect OT secretion activities

Table 5. Summary of STAI and POMS2 analysis.

Item		p value		
		Female	Male	Overall
STAI	State anxiety/P scale	0.157	0.414	0.789
	State anxiety/A scale	0.317	0.317	0.504
	State anxiety total scale	0.083	0.083	0.455
	Trait Anxiety/P Scale	0.102	1.000	0.005
	Trait Anxiety/A Scale	0.046	0.564	0.074
	Trait anxiety total scale	0.046	0.317	0.008
	5 stages/state anxiety/P scale	0.157	0.414	1.000
	5 stages/state anxiety/A scale	0.655	0.317	0.180
	5 stages/state anxiety/full scale	1.000	0.317	0.059
	5 stages/trait anxiety/P scale	0.102	1.000	0.103
	5 stages/trait anxiety/A scale	0.046	0.564	0.059
	5 levels	0.025	0.157	0.025
POMS2 (Japan version)	AH [Anger-Hostility]	0.042	0.027	0.003
	CB [Confusion-Bewilderment]	0.043	0.068	0.008
	DD [Depression-Depression]	0.046	0.027	0.003
	FI [Fatigue-Lethargy]	0.058	0.042	0.006
	TA [Tension-Anxiety]	0.102	0.058	0.012
	VA [Vitality-Vitality]	0.131	0.500	0.113
	F [Friendship]	0.038	0.465	0.083
	TMD score	0.046	0.027	0.003

Results are expressed as p-values for before-and-after comparisons by Wilcoxon signed rank test, female n = 6, male n = 6. Pink high-lighted area shows items with significant difference only for female. Gray high-lighted area shows items with significant difference only for male. STAI, State-Trait Anxiety Inventory-Form JYZ; POMS2®, Profile of Mood States 2nd Edition Japanese

Significant difference only for female

Significant difference only for male

Wilcoxon signed rank test (Before vs After 1 week)

and would contribute to the safe delivery of babies. We are eager to see further investigation conducted on this issue.

Limitation of study

It is natural to think that social contacts are related to sleep quality, as social contacts occur before and during sleep. According to public opinion surveys, in Western societies, 85% of people sleep with their partners, pets, or children[64]. When a person shares a bed with someone, it is possible that contact behaviors are facilitated before and during sleep[65]. The present trial had an inclusion criterion of "individuals who sleep alone" and in principle, there were no cases of sharing a bed. However, contact stimuli before sleep were not investigated in a background survey.

The present study did not analyze vasopressin. Changes and differences between men and women were undetected in vasopressin secretion regarding improvements of "sleep quality". A possibility has been indicated that aggressiveness would increase via vasopressin secretion[32]. This could complicate the issue. Further investigation into vasopressin secretion is warranted.

Safety endpoints

No adverse changes were recognized in this trial regarding blood pressure (systolic/diastolic), pulse rate, body weight, somatic fat rate, or BMI. No adverse events caused by the test product were observed in the subjects of the present study.

Conclusion

We conducted an open-label trial of the test product mattress (a distinctive 4-layer, 3-dimensional structure mattress: "AiR SX") with 12 male and female subjects, who could have unappropriated bedding. The test products, the bed mattresses, were used for one week and a questionnaire survey on "sleep quality" and stress-related biomarkers were assessed. Before the trial, the usage of unappropriated bedding induced mental stress. It was assumed that via increased OT and decreased NA neuron activity in the sympathetic nerve system, disturbances of homeostasis were

responded to. The usage of appropriate bedding improved "sleep quality" and stress loads were reduced. As a result, the data showed a significant increase of NA metabolite (VMA) in both men and women, a significant decrease in OT at wake-up time in women, and an increase in OT in women at bedtime. These findings suggested the possibility that stress response reactions were alleviated and OT secretion activity was regained due to favorable stimuli (*Fig. 4*). No adverse events were recognized regarding safety. It is possible that the deteriorated "sleep quality" induced by incompatible bedding, is recognized as a mental stress, leading to disturbances in the homeostasis of physical functions. It is important to select appropriate bedding and improve "sleep quality" in order to reduce mental and physiological stresses and maintain homeostasis in OT secretion.

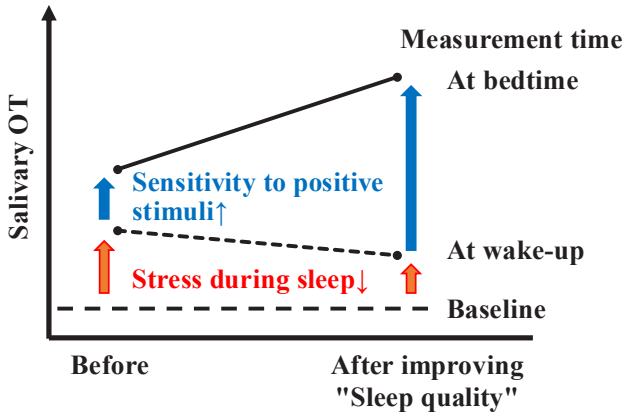

Fig. 4. **Alternation of OT secretion due to improvements in "sleep quality" (hypothesis).**

OT, oxttocin.

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Conflict of Interest

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Competitive research funding was not obtained.

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