Abstract This study investigated the physiological and psychological effects of sanitary napkins (SN) on women in hemorrhage treatment during the menstrual phase. Mesh and non-woven napkins were employed, and the effects were studied during the follicular and menstrual phases; mesh SN presented a higher textural surface-roughness. In both phases, the increases in systolic and diastolic blood pressure were significantly dependent on the application intervals. The low-frequency component of systolic blood pressure variability significantly increased, while the salivary secretion rate decreased with the use of mesh SN during the follicular phase compared with non-woven SN. In addition, the heart rate during the menstrual phase significantly increased in subjects after the replacement of mesh SN compared with non-woven SN. In cases of wearing the unpleasant mesh SN, electroencephalography (EEG) manifested bilateral enhancements in \( \beta \) and \( \alpha_2 \) waves in the frontal areas increased arousal level during both phases. From the above findings, napkin use increased physiological loading and wearing napkins with higher textural surface-roughness tended to increase activities of the autonomic nervous system and brain arousal level. J Physiol Anthropol Appl Human Sci 24 (1): 7–14, 2005 http://www.jstage.jst.go.jp/browse/jpa

Keywords: sanitary napkin, menstruation, electroencephalography, autonomic nervous activity

Introduction

The menstrual cycle, intrinsic of female physiology, persists for 3–7 days monthly over a 40-year period. As such, females have to live with menstruation for a period corresponding to 4–9 years of their lifespan. Although menstruation is very important from the viewpoint of preservation of the human species, females undergo substantial unpleasant events such as menstrual pain, premenstrual syndrome and menstrual bleeding during the menstrual cycle that require proper treatment measures to attenuate the discomfort encountered. The present study focused on the unpleasantness experienced during the menstrual phase by the application of sanitary napkins (SN). Although the unpleasantness may be induced when SN is used on humans, these physiological problems can be attenuated by technical improvements of SN based on the special features of human physiology. The unpleasantness of using SN could exacerbate physiological and psychological discomfits during the menstrual phase.

In research relating to SN, attention has recently been focused on physical factors such as absorption aspects and psychological perspectives involving skin contact or tactile sensation. Typical studies on menstruation products have revealed that poor aeration, leakage/stuffiness, rashes and vulvar irritation or contact dermatitis are induced by wearing SN (Ogawa and Kan, 1994; Eason and Felman, 1996). However, studies on physiological responses to tactile stimuli on wearing SN have hitherto not been attempted.

However, numerous studies of physiological changes accompanying the menstrual cycle have been conducted to examine the effects on autonomic nervous system (ANS) activity, Saeki et al. (1997) have demonstrated significant increases in the high-frequency component (HF) of heart rate variability (HRV), indices of parasympathetic nervous system (PNS) activity (Pomeranz et al., 1985), during the follicular phase compared with the menstrual phase. Although fluctuations in ovarian hormones are associated with the unchanged blood pressure (BP) and heart rate (HR) during the menstrual cycle, LF/HF of HRV significantly increased during the early and middle luteal phases compared with the late follicular phase (Hirshoren et al., 2002). However, according to Mehta and Chakrabarty (1993), differences in PNS activity attributed to the menstrual cycle were not significant, albeit the systolic blood pressure (SBP) was significantly elevated in the pre-menstrual phase compared with the menstrual and post-menstrual phases. In short, a common understanding of the changes in ANS activity accompanying the menstrual cycle has not been established as yet. This could have been due to various factors; e.g., seasonal differences, measurement time and others. However, there is also the need to consider the fabric used in the manufacture of SN. According to Watanuki and Mitarai (1999), rough underwear depresses ANS activity, or elicits inhibitory effects on both PNS and sympathetic nervous system (SNS) activities. As such, it can not be denied...
that SN materials employed in previous studies could have yielded the controversial outcomes.

Furthermore, as homeostasis of the human body is maintained by collective coordination of the central nervous system (CNS), ANS, endocrine system and immune system, observations of multiple systems related with physiological changes induced when using SN are warranted. As such, our present study attempted to examine the effects of wearing SN on CNS, ANS and the immune system in human subjects.

**Methods**

**Subjects**

Subjects were 12 female undergraduates (mean age: 21.31 ±0.95 years) with regular menstrual cycles. The subjects in the experiment put on similar attire such as sanitary briefs, T-shirts and short pants but did not wear brassieres. Written informed consent was obtained from all volunteers after a full explanation of the experimental purpose and protocol. All subjects were in good health, not taking any prescription medication including oral contraceptives or psychotropic agents, and of normotensive BP status. Subjects were asked to abstain from eating, drinking, smoking and exercise for at least three hours before the experiment.

**Materials**

Two current mainstream SN types were employed in this study; mesh and non-woven. The infiltration speed of non-woven SN was 3 folds that of mesh SN. Although the textural surface-roughness of non-woven SN was low, reflux of discharges to the surface facilitated infiltration. This phenomenal reflux, or rewet, was less in mesh than non-woven SN; however, the former displayed higher textural surface-roughness (Table 1).

**Experimental design**

Experiment I was conducted during the follicular phase (days 8–11 after menstruation). To investigate the unpleasantness induced by wearing SN, it was necessary for us to first examine the different effects on the human body induced by differences in the physical features of SN surface imposed on the skin due to friction generated from SN-skin contact. The follicular phase was preferred to the menstrual phase presents a better physiologically stable state than the luteal phase, thus facilitating execution of the experimental protocol.

To investigate the effects of different SN fabrics on menstrual-phase physiological responses, Experiment II was conducted during the menstrual phase (days 3–5 after menstruation). SN were distributed to subjects before menstruation onset, and they were instructed to wear the given SN upon menstrual initiation. Experiment I and II were conducted in a room maintained at 27±1°C with relative humidity of 50±10%.

**Measurements**

In Experiments I and II, electrocardiography (ECG), BP, EEG and subjective evaluations were monitored at rest. ECG measurement (NEC, EE5514) was performed by synchronizing respiratory systemic control at 0.25-Hz metronorm for 180 sec. From the ECG monitoring, HR and HRV were derived. Many studies have reported the reliability of HRV as a noninvasive index of ANS activity (Ishibashi et al., 1999; Kobayashi et al., 1999; Miyatsuji et al., 2002). HF of HRV relates to parasympathetic activity and the low frequencies (LF) of HRV are mediated by both parasympathetic and sympathetic activities (Pomeranz et al., 1985). And the LF/HF ratio was proposed as an index of sympatho-vagal balance by Pagani et al. (1986). Moreover, the BP levels were consistently being monitored. The values of LF of SBP variability (SBPV), which reflect the sympathetic nervous system of the vasomotor nerve (Oka et al., 1992; Mukai et al., 1994; Okuda et al., 2002), were evaluated as well.

As an increase in secretory immunoglobulin A (s-IgA) implies a defense response of the body system, especially reactions in response to the stress from the immune system, quantitative evaluations of s-IgA have recently been extensively employed as an important index in measuring stress response (Zeier et al., 1996; Willemsen et al., 1998). From the ECG monitoring, HR and HRV were derived. Many studies have reported the reliability of HRV as a noninvasive index of ANS activity (Ishibashi et al., 1999; Kobayashi et al., 1999; Miyatsuji et al., 2002). HF of HRV relates to parasympathetic activity and the low frequencies (LF) of HRV are mediated by both parasympathetic and sympathetic activities (Pomeranz et al., 1985). And the LF/HF ratio was proposed as an index of sympatho-vagal balance by Pagani et al. (1986). Moreover, the BP levels were consistently being monitored. The values of LF of SBP variability (SBPV), which reflect the sympathetic nervous system of the vasomotor nerve (Oka et al., 1992; Mukai et al., 1994; Okuda et al., 2002), were evaluated as well.

Subjective evaluations of unpleasantness were scored to the nearest 0.5 as follows: very unpleasant (−2); unpleasant (−1); indifferent (0); pleasant (1); and very pleasant (2).

**Procedures**

The experimental procedures (Fig. 1) indicated that subjects were first seated in a resting state for 20 min without SN

<table>
<thead>
<tr>
<th>Size (mm)</th>
<th>Weight (g)</th>
<th>Infiltration speed (sec)</th>
<th>Rewet (g/5 min)</th>
<th>Frictional coefficient</th>
<th>Mean deviation of the frictional coefficient</th>
</tr>
</thead>
<tbody>
<tr>
<td>Non-Woven</td>
<td>82×210</td>
<td>6.0</td>
<td>6.7</td>
<td>2.0</td>
<td>0.142</td>
</tr>
<tr>
<td>Mesh</td>
<td>85×210</td>
<td>6.0</td>
<td>18.3</td>
<td>1.0</td>
<td>0.312</td>
</tr>
</tbody>
</table>
application followed by a 40-min resting period with SN application in Experiment I (follicular phase). Objective indexes such as EEG, ECG, BP and subjective evaluations were monitored at 10-min intervals after SN application. Saliva sampling was performed before and 35 min after SN application.

In Experiment II (menstrual phase), subjects were asked to wear the initially applied SN for 20 min in a resting state, and remained in a similar resting state for another 35 min after SN replacement. At 10, 20 and 30 min after SN replacement, objective indexes such as EEG, ECG, BP and subjective evaluations were measured accordingly. Saliva sampling was conducted before and 35 min after SN replacement.

Data analysis
The ECG data were digitized with a sampling frequency of 1 KHz on a personal computer equipped with a 12-bit analog-to-digital converter (MICROSCIENCE ADM-5298BPC). The heart period sequences, obtained by detecting the peak of R wave in ECG, were converted into beats/min and interpolated into 10 Hz equidistant data. Spectral analysis of HR V was applied to the time series data of R-R intervals for every 3 minutes, by fast Fourier transformation (FFT) using a Hamming window. The LF and HF components were integrated from 0.05 to 0.15 Hz and 0.15 to 0.35 Hz of the power spectra, respectively.

We continuously monitored BP using the Finapres (Ohmeda, 2300 Finapres) with a digit cuff applied around the left middle finger and the hand supported comfortably by the subject’s side, at heart level. This noninvasive BP monitor uses the vascular unloading technique to determine SBP and DBP on a beat-by-beat basis.

EEG (NEC Medical EE5514; NEC Co. Ltd.) was recorded using an electrocap (Electro-Cap International, Inc.) from 13 sites according to the International 10-20 Electrode System (Fp1, Fp2, C3, C4, O1, O2, F7, F8, T5, T6, Fz, Cz, Pz) for 1 min with the subject seated and eye closed. Data input executed with NEC Medical EE5514 (SYNAFIT 5000) software was analyzed with BIMUTAS II (Kissei Comtec Co. Ltd.). Signal bandpass was 0.5–60.0 Hz and the digital sampling frequency was 200 Hz. Power spectra were calculated by FFT using a Hamming window. After calculating the power spectra, the rates of respective wavelength components were derived. The frequency bands were categorized as follows: θ1 (4–6 Hz), θ2 (6–8 Hz), α1 (8–10 Hz), α2 (10–13 Hz), β (13–30 Hz).

s-IgA in saliva measurements
The salivary glands comprise 3 major glands (the subaural, submandibular, and sublingual glands) besides numerous minor ones. In investigating the s-IgA level in saliva, we collected samples using 2 pieces of Salivette (Sarstedt Ltd.) at the sublingual site and 2 other pieces at sites between the inner cheek and teeth at 5-min intervals. After collection, saliva was extracted from the cotton by centrifugation (KUBOTA 2700) at 3.0×10^4 rpm for 15 min before storage at –30°C. The concentration of s-IgA in saliva was determined by enzyme-linked immunosorbent assay (ELISA). Saliva aliquots (20μl) were assayed at a dilution of 1:1,000. The coating antibody was antihuman secretory component (2μg/ml) at 100μl per well. After incubation for 2 h at 37°C, wells were washed with wash buffer (NaCl 16 g, KCl 0.4 g, Na_2HPO_4 2.3 g, KH_2PO_4 0.4 g, Tween 20 1ml, 1999 ml of deionized water). Assays were performed in triplicate against a range, 0–1,000 ng/ml, of standard Human IgA. A reference sample was incorporated into each plate. After incubation for 2 h at 37°C, wells were washed with wash buffer. For detection, an HRP-conjugated goat antihuman IgA (1:2,000) was used, directed against human IgA heavy chain. Incubation wells were filled with substrate solution (0.2M Na_2HPO_4 soln, 0.1M citric acid, ABTS, 30% H_2O_2) and incubated for 0.5 h. The reaction was stopped with NaF soln (1.25%). Developed color was
measured in a Microplate Reader (BIO-RAD Model 550). Each 5-min sample provided a measure of the saliva s-IgA concentration (µg/ml) and saliva volume (ml/min). The s-IgA secretion rate (µg/min) was calculated from the formula
\[
\text{secretion rate} = \frac{\text{concentration}}{\text{volume}}.
\]

**Statistical analysis**

All data was indicated using mean values and standard deviation (SD). HR, HRV, BP, LF of SBPV, EEG, subjective evaluations were analyzed by a two-way ANOVA with repeated measures. The factors were fabric and period. Post-hoc comparisons of means were done by using the Fisher’s protected least significant difference. Statistical analysis was performed on a personal computer using Statview 5.0J (SAS Institute Inc.), and differences with a probability value of less than 0.05 were considered significant. Changes of s-IgA after SN application were statistically verified by the paired t-test.

**Results**

**Experiment I (follicular phase)**

Results of ANOVA revealed that the following exhibited the main effects of periods: SBP (F(3, 33)=4.48; p<0.01), DBP (F(3, 33)=7.96; p<0.01) during the follicular phase. Post-hoc multiple comparisons showed that SBP (Fig. 2A) and DBP (Fig. 2B) significantly increased at 20, 30 and 40 min after application compared with 10 min after wearing SN (p<0.01). Significant effects were not established in HR, LF and HF components or LF/HF ratio of HRV. A main effect of fabric (F(1, 11)=5.48; p<0.05) was procured in the LF component of SBPV. The LF component of SBPV in cases of wearing the mesh SN was significantly higher (p<0.05) than those using non-woven SN (Fig. 3).

To study the effects of mechanical stimulation on CNS by wearing SN, EEG tracings at 10 min were compared with those at 40 min after applying SN in the follicular phase. Results of ANOVA revealed that both β and α2 activities were significantly increased with mesh SN at 40 min than at 10 min after application in the follicular phase (Fig. 4A and Fig. 4B), whereas differences in the EEG changes with non-woven SN were insignificant. A significant effect of fabric also emerged for α2 band of F7(F(1,11)=5.57; p<0.05). α2 activity of F7 exhibited significant increases in mesh SN compared with non-woven SN (Fig. 5). Significant effects of the fabrics were obtained in the α2 band of C4(F(1,11)=14.50; p<0.01), F8(F(1,11)=5.23; p<0.05), Fz(F(1,11)=8.19; p<0.05), Cz(F(1,11)=10.05; p<0.01), Pz(F(1,11)=16.61; p<0.01). α2 activities of C4, F8, Fz, Cz and Pz in cases of wearing the mesh SN were significantly lower than those in cases of using non-woven SN (Fig. 5).

The saliva of three subjects who were not able to yield the volume required for analysis was excluded from analysis. Note that changes in the salivary secretion rate were compared between SN materials as individual differences in the quantitative aspect were substantial. No significant differences in secretion rate and concentration of s-IgA during the
follicular phases were observed, although there was a significant difference in the salivary secretion rate between the fabrics ($t(16) = 2.59; p < 0.05$). The salivary secretion rate of cases wearing mesh SN was less than those using non-woven SN (Fig. 6A).

A significant main effect of fabric was observed in the form of unpleasantness ($F(1, 11) = 7.09; p < 0.05$). The levels of unpleasantness derived from wearing mesh material were significantly higher than the nonwoven fabric (Fig. 6B).

The saliva of three subjects who were not able to yield the volume required for analysis was excluded from analysis. No significant differences in the s-IgA secretion rate, s-IgA concentration and salivary secretion rate were observed. No significant effects emerged for unpleasantness in the menstrual phase.

**Discussion**

**SN application-induced physiological loads**

The influence of SN use on the human body was investigated. During the follicular phase, both mesh and non-woven SN elevated the ANS-related SBP and DBP values (Fig. 2A, 2B). These increases were initiated 20 min after SN application and persisted until 40 min after application. In addition, the persistent increases of SBP and DBP were encountered from 20 min after SN replacement (Fig. 7A, 7B) even in the menstrual phase. In general, various physiological responses are triggered as a global defense mechanism of the
human body in response to certain physical stimuli assessed as stressors by the brain. The increase in BP is a typical stress response (Williams, 1986; Sawada et al., 1997). According to Williams (1986), hemodynamic reactions triggered by stress stimuli may assume 2 major patterns; viz., active coping (i.e., pattern I) and passive coping (i.e., pattern II). While Pattern I depicts an encounter with a potent stressor with active coping of a fight-or-flight response to overwhelm an emergency state, Pattern II assumes a state of monitoring unpleasantness via passive coping with inhibitory movements. In this study, the BP increase reflected the mechanical stimulus induced by SN application as a passive stressor to the human body.

Generally, when the human body manifests stress responses,
Differences in the physiological responses between the SN materials

In the follicular phase, EEG activations were induced by mesh SN application, and such phenomenal responses were not incited with use of the non-woven SN (Fig. 4A, 4B). This suggests the difference of SN material in exerted different physiological loads on application. Even during the menstrual phase, relative output of $\alpha 2$ wave in brain sites Fp1, Fp2, F8, Fz, Pz and O1 were significantly higher with mesh SN than with non-woven SN (Fig. 9). Based on EEG findings in the present study, the frontal brain sites manifested bilaterally enhanced activities with mesh SN in both follicular and menstrual phases. According to Brauchli et al. (1995), offensive odors that induced unpleasantness evoke increases of $\alpha 2$ (9.75-12.5 Hz) waves in bilateral frontal and parietal sites of brain. Moreover, Kim and Watanuki (2003) have observed enhanced extensive brain activities in frontal and other sites (Fp1, Fp2, F3, F7, F8, C4, T4, T6, P4, O1, O2) in subjects exposed to unpleasant odors. In the present investigation, although the stimulation was different from olfactory stimuli described above, the fact that significant increases in $\beta$ and $\alpha 2$ waves evoked in bilateral frontal sites on wearing mesh SN coincide with responses observed with exposure to unpleasant odors implies that the brain responses (unpleasantness) were convergent; mesh SN incited a response more unpleasant than that induced by nonwoven SN (Fig. 6B). Even when the stimuli were different, psychologically unpleasant responses might bilaterally activate frontal brain activities when viewed with EEG.

Differences of physiological responses between SN are not only reflected in EEG, changes in ANS activities were apparent as well. In the follicular phase, the LF component of SBPV with mesh SN indicated significantly (p<0.05) higher values than those with non-woven SN (Fig. 3). As Oka et al. (1992) have demonstrated significant correlations of BP increases with the LF component of SBPV in the valsalva overshoot and cold pressor test, the LF component of SBPV is thus closely associated with $\alpha$-sympathetic functions of the vasomotor nerve. In short, use of mesh SN in the follicular phase is more likely to enhance vasomotor sympathetic activity, and eventually increases peripheral resistance as compared with non-woven SN. Although no effects on the LF component of SBPV were encountered in the menstrual phase, a main effect of fabric in HR was noted. In other words, mesh SN markedly (p<0.05) elevated cardiac sympathetic nerve activity (as reflected by the increases in HR) compared with non-woven SN (Fig. 8). Increases in both LF of SBPV and HR contributed to elevations in blood pressure, a physiological response that is undesirable for the human body. Although a difference in BP was not induced with different SN materials in the present study, the fact that substantial increases in HR and the LF component of SBPV attributed to BP increases with mesh SN suggests that mesh SN were more likely to enhance SNS activity than non-woven SN. Investigations of this perspective included inferences from the measurements of salivary secretion as well (Fig. 6A). An increase in SNS activity is generally reciprocated by attenuation in PNS activity. Salivary secretion is depressed when PNS activity is inhibited (Okura et al., 2000; Tanaka and Midorikawa, 2003). In the follicular phase, as the use of mesh SN quantitatively reduced salivary secretion rate more than the use of non-woven SN, wearing mesh SN probably elevated SNS activity.

From the present findings, mesh materials are more likely to elevate SNS activity than non-woven materials. Although Hirshoren et al. (2002) have reported no changes in BP and HR during the menstrual cycle, the outcome may be determined by the SN material worn in the menstrual phase. It is thus critical to obtain accurate quantitative evaluations with different SN materials during the menstrual phase.

Based on the present results, wearing SN enhanced physiological loading, and SN made of mesh materials, or textures with higher roughness, were more likely to induce a higher degree than those made of non-woven materials or textures with a lower roughness. The use of SN with textures of a high roughness tends to enhance physiological loading and exacerbate the various physiological symptoms experienced during the menstrual phase. In other words, it is possible to reduce unpleasant physiological loads with improved SN materials. Furthermore, our findings suggest that accurate results are more likely to be procured when participants are subjected to wearing similar SN in cases that physiological responses are investigated during the menstrual phase.

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