ORIGINAL ARTICLE

Verification of endocrinological functions at a short distance between parametric speakers and the human body

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Abstract

In recent years, a new type of speaker called the parametric speaker has been used to generate highly directional sound, and these speakers are now commercially available. In our previous study, we verified that the burden of the parametric speaker was lower than that of the general speaker for endocrine functions. However, nothing has yet been demonstrated about the effects of the shorter distance than 2.6 m between parametric speakers and the human body. Therefore, we investigated the distance effect on endocrinological function and subjective evaluation.

Nine male subjects participated in this study. They completed three consecutive sessions: a 20-min quiet period as a baseline, a 30-min mental task period with general speakers or parametric speakers, and a 20-min recovery period. We measured salivary cortisol and chromogranin A (CgA) concentrations. Furthermore, subjects took the Kwansei-gakuin Sleepiness Scale (KSS) test before and after the task and also a sound quality evaluation test after it. Four experiments, one with a speaker condition (general speaker and parametric speaker), the other with a distance condition (0.3 m and 1.0 m), were conducted, respectively, at the same time of day on separate days. We used three-way repeated measures ANOVA (speaker factor × distance factor × time factor) to examine the effects of the parametric speaker.

We found that the endocrinological functions were not significantly different between the speaker condition and the distance condition.

The results also showed that the physiological burdens increased with progress in time independent of the speaker condition and distance condition.

Introduction

In recent years, a new type of speaker called the parametric speaker has been used to generate highly directional sound, and these speakers are now commercially available. The parametric speaker is a sound system that can maintain a very sharp directivity by using an ultrasonic wave. These high-directional speakers based on parametric arrays have allowed sound transmission within a narrow range of acoustic space like a 'spotlight' (Komatsuzaki *et al* 2010). In addition, with the parametric speaker it is difficult to recognize the distance to the sound source because of the lack of reverberant sound.

In our previous study, we measured the effects of parametric sound on humans with the speakers set at a distance of 2.6 m from the subjects (Lee *et al* 2010 and 2011).In that study we verified that the parametric speaker sound resulted in a lower stress level than that generated by the general speaker sound with regard to the endocrinological system, especially with regard to salivary cortisol. However, nothing has been revealed about the effect of the parametric speaker sound on endocrinological responses as a result of a shorter distance than 2.6 m between these speakers and the subject, until now.

Recently, several studies have demonstrated that the secretion of salivary stress biomarkers such as salivary cortisol and salivary chromogranin A (CgA) increased under different stress conditions (Takai *et al* 2004; Miyakawa *et al* 2006; Nater *et al* 2006; Hebert &Lupien 2007; Hellhammer *et al* 2009). In particular, levels of stress hormones (i.e., catecholamine and cortisol) have been used to evaluate the physiological effects of noise (Babisch 2002). In the previous studies, one proposed biological mechanism is that noise causes a release of stress hormones, which in turn adversely affect cardiovascular risk factors (Babisch *et al* 2001; Ising & Kruppa 2004; Spreng 2000) and myocardial infarction (Jarup *et al* 2008). In addition, an intermediary mechanism may involve the metabolic syndrome, in which a disturbed hypothalamus-pituitary-adrenal (HPA) axis regulation has been assumed to play an important role (Chandola *et al* 2006).

Salivary cortisol is often used as a stress index because its measurement has various advantages, such as a non-invasive collection procedure. The cortisol is the principal hormonal product of the human HPA axis, which is a close-looped endocrine system. The glucocorticoid hormone cortisol is the main secretory product of the neuroendocrine cascade and a valid indicator of stress (Evans *et al* 2001; Miki *et al* 1998; Schulz *et al* 1998). In addition, acute and chronic stress has been reported to increase the activity of the HPA axis, with a subsequent rise in cortisol concentrations (Al'Absi *et al*1997: Vedhara *et al* 1999). Furthermore, cortisol has recently been suggested to be a useful parameter to measure noise-related stress (Bigert *et al* 2005). After long-time stressful noise exposure, the ability to down-regulate cortisol may be inhibited (Spreng 2000). In previous studies, increased cortisol concentrations were reported during the anticipation of stressful experiences such as public speaking (Basset *et al* 1987), academic examinations (Maes *et al* 1998), and dental procedures (Miller *et al* 1995). On the other hand, the catecholamine levels serve as an objective index of stress because the sympathetic-adrenomedullary system responds to stress (Lee *et al* 2006). The CgA level may serve as a potential non-invasive tool for evaluating the sympathetic nervous system following psychological stress (Walsh *et al* 1999). Furthermore, a rapid and sensitive elevation of salivary CgA was reported in response to psychosomatic stressors such as a public speaking and driving a vehicle (Nakane 1998 and 2002).

The purpose of the present study was to investigate the influence of a shorter distance than 2.6 m between parametric speakers and the subjects on endocrinological responses using the salivary hormones. In addition, we carried out a subjective evaluation of the parametric speakers.

Tab. 1. The physical characteristic of the subjects.

Subjects	Age (yr)	Height(cm)	Weight(kg)	BMI
Sub 1	25	165	65	23.9
Sub 2	23	177	62	19.8
Sub 3	22	171	57	19.5
Sub 4	23	172	69	23.3
Sub 5	24	165	47	17.3
Sub 6	24	178	71	22.4
Sub 7	23	184	70	20.7
Sub 8	24	172	54	18.3
Sub 9	22	173	65	21.7
Mean±SD	**23±1.0**	**173±6.08**	**62±8.1**	**20.8±2.26**

Methods

Subjects

Nine healthy male students (23±1.0 years, 173±6.08 cm, 62±8.1 kg) participated in this study.

This study was designed to eliminate many of the variables known to cause alterations in salivary cortisol (Perna *et al* 1997; Hooper *et al* 1993). Subjects were instructed to refrain from any intense physical activity, dietary supplements, and medicines for 24 hours before the experiment session. Furthermore, sex, tobacco, alcohol, and caffeine consumption were restricted for 12 hours before the experiment day. Subjects were asked to maintain their regular sleep-wake cycle.

The subjects performed an auditory test (ITERA, GN Otometrics) before the experiment. Their hearing ability was confirmed to be normal. Informed consent for participation in the study, approved by the bioethics committee of the Graduate School of Engineering, Chiba University, was obtained from all subjects. Their physical characteristics are shown in ***Table 1.***

Protocol

The experiments were conducted in a soundproof room. Four experiments were conducted at the same time of day on separate days and under the same conditions with the exception of the speaker condition (general or parametric speaker) and the distance condition (0.3 m and 1.0 m between speakers and subjects). Subjects were asked to relax for at least 15 min after they arrived at the soundproof room. Subjects completed three consecutive sessions: a 20-min quiet period as a baseline, a 30-min mental task period with the general speaker or parametric speaker, and a 20-min recovery period. Subjects were told to rest and physically relax throughout the experimental period. The experimental protocol is shown in ***Figure 1***. The order of the four conditions was counterbalanced between the subjects. A salivary sample was taken four times, i.e., before the task, immediately after the task, 10 min after the task, and 20 min after the task.

Analysis of salivary stress biomarkers

In a previous study, it was verified that cotton Salivettes interfered with cortisol concentration and that cotton-based devices artificially reduced free cortisol concentration (Gröschl *et al* 2006 and 2008). Therefore, in collecting salivary cortisol samples, we did not use Salivettes but instead employed plastic tubes. Before saliva collection, subjects were required to rinse out their mouths for one minute with water to remove any substances such as chlorine that may affect cortisol (Hooper *et al* 1993). Immediately after sampling, samples were frozen and stored at –28 °C until they were assayed for cortisol and CgA concentrations. Samples were later thawed and centrifuged for 15 min at 3 000 rpm and processed according the manufacturer's instructions. The cortisol was determined by EIA especially designed for the assay of cortisol in saliva (Salimetrics, USA). The cortisol was expressed as μg/dL. In addition, the CgA concentration was used as the salivary Chromogranin A kit (YK070 Human Chromogranin A EIA, Japan) in the analysis. We determined the saliva CgA concentration by correcting the value of the total salivary protein measured in the sample. Values of CgA were calculated based on values of total salivary protein measured in the same sample. The concentration of protein in the saliva was based on the Bradford method. CgA activity was expressed as pmol/mg. All samples were assayed in duplicate.

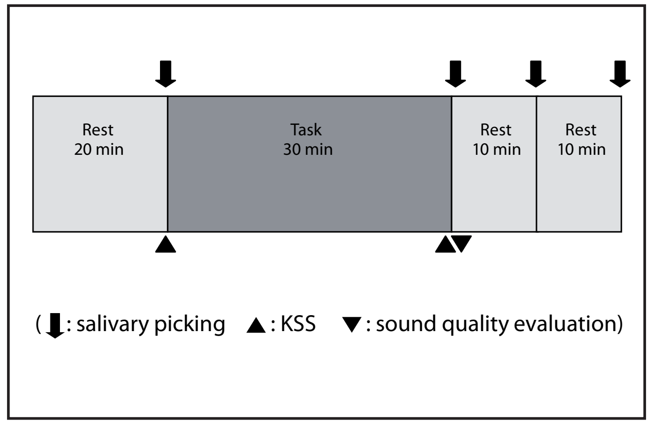

Fig. 1. The protocol of the experiments.

Mental task

The mental task consisted of normal sentences and deviant sentences coming from the speaker. Subjects were instructed to judge whether a sentence was true or false and to push a button to indicate that as quickly as possible. The sentences (in Japanese) were made by a text-to-speech synthesis software (SMART TALK Version 3, OKI). For example, a normal sentence was "watasi ha ringo wo taberu" (I eat an apple). In this case, the subjects were asked to push a 'red' button. By contrast, a deviant sentence was "watasi ha ringo wo nomu" (I drink an apple). In this case, the subjects were asked to push a 'blue' button. The Leq of the sound generated by the general speaker was 72.4 dBA, and the Leq of sound generated by the parametric speaker was 72.3 dBA. The background Leq of the soundproof room

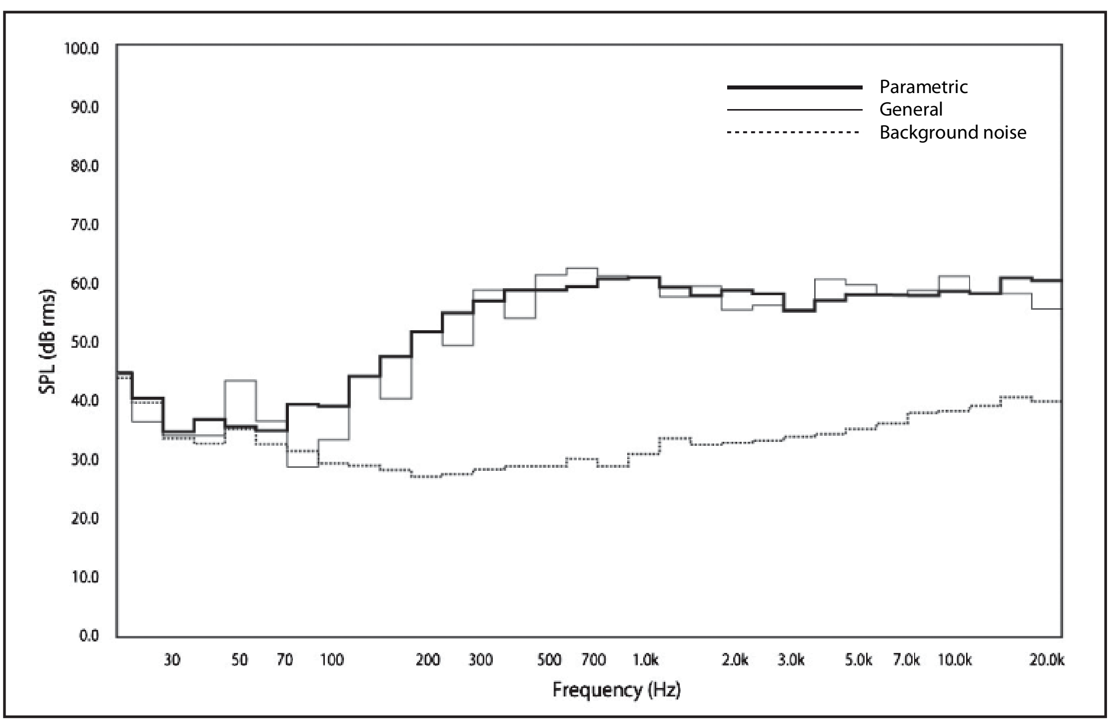

Fig. 2. The frequency characteristic of the general speaker and parametric speaker

was 52.2 dBA. The output characteristics of the general speaker and the parametric speaker were almost the same. ***Figure 2*** shows the frequency characteristic of the general speaker and parametric speaker.

Questionnaire for the sound quality evaluation and KSS

For the sound quality evaluation, the subjects were asked to record their responses with the visual analogue scale (VAS). The Kwansei-gakuin Sleepiness Scale (KSS) was used to assess subjective sleepiness. Value on the scale concerning drowsiness was determined from answers to 22 questions. The higher the total, the more drowsy the subjects felt.

Tab. 2. The sensation of sound quality evaluation.

The sound quality evaluation sensations	1.0 m or less		2.6 m	
	parametric	general	parametric	general
Hardness	5.583	4.850*	5.777	4.844
Annoyance	6.056	4.117	5.177	4.422
Tune	3.533	3.267	4.933	5.233
Volume of sound	4.917	4.569	4.933	5.233
Warm	3.411	3.281+	3.233	5.011+
Comfort	3.606	4.483*	4.111	4.666
Noisiness	3.328	3.122	3.955	3.111+
Clearness	2.772	3.861**	3.766	4.544
Mellifluence	4.139	4.367	3.988	4.733
Audible	4.506	4.767	3.933	4.977
Information collecting ability	4.428	3.978	4.411	4.522

(+$p<0.1$; *$p<0.05$; **$p<0.01$)

Statistical analyses

We calculated the mean for all indexes. In the salivary hormones, changes (Δ) calculated by subtracting the respective baseline values from the average values were used to conduct statistical analyses. For the cortisol, CgA and subjective evaluations, a three-way repeated-measures ANOVA (speaker factor × distance × time factor) was conducted. When a significant F value was found, we performed a Bonferroni test as a post-hoc test. All statistical analyses were performed using SPSS 11.0J (SPSS, Japan). Differences with values of $p<0.05$ were considered significant. Data are shown as the mean ± standard error of the mean unless otherwise stated.

Results

Figure 3 shows the changes of cortisol concentration in the speaker condition and distance condition. The main effect of the speaker factor [$F(1,8)=0.194$, $p=0.671$] and distance factor [$F(1,8)=0.639$, $p=0.447$] was not significant. However, the main effect of the time factor was significant [$F(1,8)=8.752$, $p=0.0182$]. The changes of cortisol concentration during the task period were significantly smaller than those during the recovery period. Meanwhile, the CgA concentration was not significantly different between the speaker factor [$F(1,8)=1.736$, $p=0.224$], distance factor [$F(1,8)=0.174$, $p=0.687$], and time factor [$F(1,8)=1.613$, $p=0.298$] (***Figure 4***). Some subjects showed a prolonged elevation in their CgA levels; others showed either immediate recovery or no effects.

For the "Hardness" sensation from the sound quality evaluation, the score of the parametric speaker was significantly higher than that of the general speaker [$F(1,8)=10.257$, $p=0.012$]. For the "Comfort" [$F(1,8)=20.317$, $p=0.002$] and "Clearness" [$F(1,8)=16.096$, $p=0.003$] sensations, the score of the parametric speaker was significantly lower than that

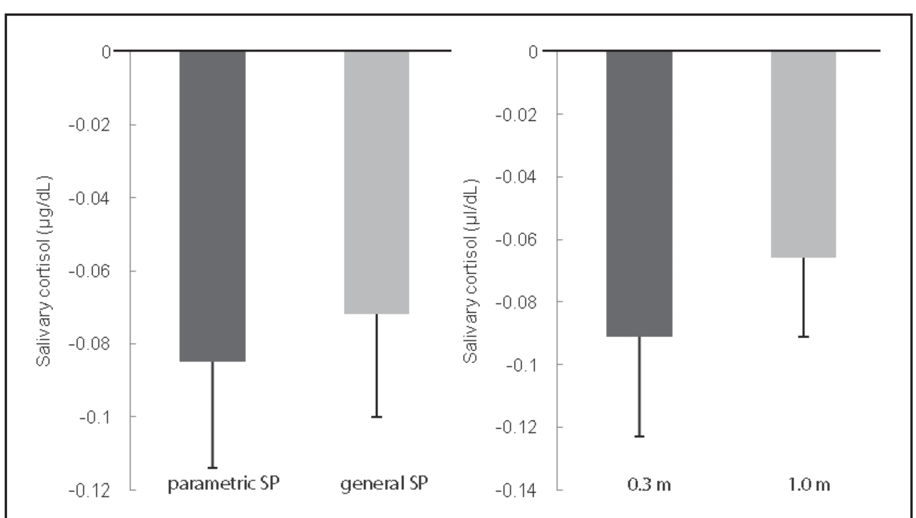

Fig. 3. The changes of cortsiol concentration in the speaker condition (left) and distance condition (right).

of the general speaker. In the result from the KSS test, subjects listening to the parametric speaker tended to have a higher KSS score than they did when listening to the general speaker. ***Table 2*** shows the results of the 11 items for sound quality evaluation are shown in comparison to results of a previous study at 2.6 m distance.

Discussion

In our previous studies, the effects of parametric speaker sound on salivary cortisol and CgA concentrations, especially in the endocrinological system, were demonstrated (Lee *et al* 2010). Furthermore, we showed that the sound of the parametric speaker set 2.6 m from the subjects was less stressful than that of the general speaker with regard to the cardiovascular system (Lee *et al* 2011). From these studies, we determined the characteristics of parametric speakers compared to general speakers at a distance of 2.6 m. However, nothing had previously been demonstrated about the effect of the sound of parametric speakers at shorter distances from the subjects. Therefore, we measured the endocrinological responses to the sound by general speaker and parametric speaker in shorter distance conditions (0.3 m and 1.0 m) in the present study.

The main finding of this study was that the endocrinological effects of the parametric speaker were not obtained at the distance of 1 m or less. To be more specific, a three-way repeated measure ANOVA on cortisol concentration showed no significant effects on the speaker factor or the distance factor. By contrast, a significantly higher salivary cortisol concentration was observed with progress in time.

We observed no effects of sound of the parametric speaker on salivary cortisol or CgA in our subjects in the present study. There may be several reasons for this. The first reason is related to the characteristics of salivary cortisol and CgA under stress. First of all, acute activation of the HPA axis also represents major physiological responses to environmental stressors (Waye *et al* 2003). In particular, the effect of noise on cortisol concentrations in the HPA axis has been evaluated. For instance, Hébert et al (2005) reported cortisol elevation when workers were exposed to techno-music (85 dBA) for 16 hours. In addition, previous studies on urine cortisol have found increased levels of urine cortisol after chronic exposure to transportation noise (Evans *et al* 2001). However, the results in the present study did not correspond with the results of these reports. We suspect that cortisol response is related to stress intensity, exposure time, and stress type. According to Melamed and Bruhis (1996) and Herbert and Lupien (2009), in humans and animals increased cortisol was found following noise exposure of longer duration, higher noise levels, or both. However, the Leq of parametric speakers and general speakers in the present study (72.3 dBA and 72.4dBA, respectively) were lower than those of the previous study (85 dBA and 95 dBA, respectively) (Melamed & Bruhis 1996). Furthermore the noise exposure time in our study was shorter than that in the previous studies (Melamed & Bruhis 1996; Herbert & Lupien 2009). The stress caused by the noise exposure in the present study may not have been intensive. In other words, cortisol levels change in response to intensive stress but not to continuous and relatively low stress. As another possibility, there may be individual differences in cortisol levels. It was reported that a number of factors are known to influence HPA axis function, including a history of early life stress (Carpenter *et al* 2007; Gunnar & Vazquez 2001; Heim *et al* 2000; Shea *et al* 2005) as well as mood and anxiety disorders. Thus, the difference in the cortisol concentration in response to the parametric speaker and general speaker might not have appeared.

Catecholamine levels are also commonly used as a sensitive biochemical index of stress; however, it is dif-

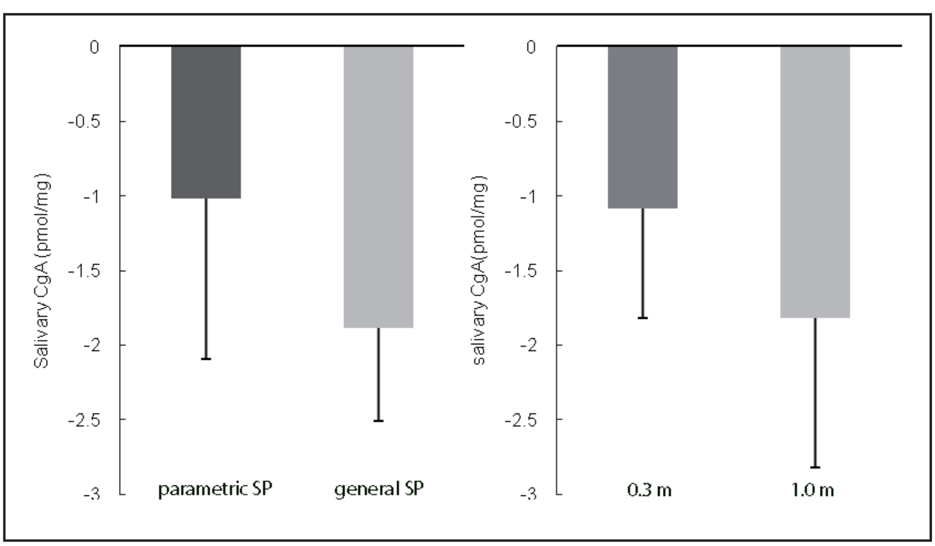

Fig. 4. The changes of CgA concentration in the speaker condition (left) and distance condition (right).

ficult to measure their concentrations in the saliva due to generally low levels and rapid degradation. Therefore, salivary CgA is used as a substitute marker for catecholamines, as it reflects psychological stress more quickly and sensitively than does cortisol (Nakane 1998 and 2002). In this study, we observed no difference of the CgA levels in response to the general speaker and the parametric speaker. We suppose that the reason may be related to the individual differences and the characteristics of the CgA as measured by the ELISA. According to Miyakawa *et al* (2006), large individual differences have been found in changes in salivary CgA levels in responses to noise exposure. Some subjects in the present study showed a pronged elevation in CgA levels, while others showed either immediate recovery or no effects. It seems reasonable to suppose that these individual differences correlate with different physiological sensitivities to noise. In addition, the difference seems to be caused by the test-specific (ELISA) wide range of the CgA concentrations obtained from saliva samples collected after sound exposure. Another reason for such an inhomogeneous distribution of results may be due to the poor stability of this metabolite in the sample matrix (Bender *et al* 1992). Moreover, measuring CgA using the ELISA technique is strongly investigator dependent and is laborious compared to salivary α-amylase determination (Wagner *et al* 2010).

The second reason that our results may differ from those of earlier studies is a characteristic of parametric speakers. In several previous studies, it has been reported that the sound focusing of a parametric speaker (Komatsuzaki *et al* 2010) could be utilized to deliver audible information to people in a particular region without disturbing others (Ju *et al* 2010) because the directivity was so strong (Croft & Norris 2001). In an earlier study, Nabelek *et al* (1989) verified that the articulation of sounds was decreased by reverberation. Therefore, we supposed that a parametric speaker has less reverberation and higher articulation because of its strong directivity. When speakers are set a certain distance from subjects, we can presume that a general speaker has more reverberation and lower articulation compared to a parametric speaker. This fact was demonstrated in our previous study (Lee *et al* 2011). However, we estimated that there was little difference in articulation between the parametric and general speaker sounds at the shorter distance. At the shorter distance, it is possible that the subjects perceived that the general speaker sound was nearly equal to the parametric speaker sound, because the reverberation of the general speaker was decreased. Therefore, we considered that there was little effect of the directional characteristic of the parametric speaker at distances of 1 m or less. As a result, the speaker condition might not have caused any endocrinological difference.

In conclusion, the main finding of this study was that the endocrinological effects of the parametric speaker were not obtained at the distance of 1 m or less. We believe this supports the findings of our previous study from the viewpoint of endocrinology, and this information will be important in future studies when considering applications of parametric speaker sound.

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