

# Factors Affecting and Adjustments for Sex Differences in Current Perception Threshold With Transcutaneous Electrical Stimulation in Healthy Subjects

Shin-ichiro Seno, PhD\*<sup>†</sup>; Hideaki Shimazu, PhD<sup>†</sup>; Eiki Kogure, PhD\*<sup>†</sup>; Atsushi Watanabe, PhD\*<sup>†</sup>; Hiroko Kobayashi, PhD\*

**Objective:** Current perception threshold (CPT) measurement is a noninvasive, easy, and semi-objective method for determining sensory function using transcutaneous electrical stimulation. Previous studies have shown that CPT is determined by physical characteristics, such as sex, age, physical sites, and presence of neuropathy. Although the CPT reported in males is clearly higher than that in females, the reason for this difference remains unclear. This study investigates the cause of sex-based differences in CPT and suggests an adjustment method, which may suppress the sex difference in CPT.

**Materials and Methods:** Electrical stimulation was applied with PainVision<sup>®</sup> via five sizes of circular surface electrodes. Seventy healthy participants were examined thrice under each electrode. The relationship among body water percentage, body fat percentage, and CPT was then analyzed.

**Results:** CPT values are higher in males than that in females, with statistically significant sex differences with each electrode pairs (EL 1:  $p < 0.001$ ; EL 2:  $p = 0.006$ ; EL 3:  $p < 0.001$ ; EL 4:  $p < 0.001$ ; EL 5:  $p < 0.001$ ). By adjusting for body fat percentage or body water percentage, the log-transformation values (CPT values) no longer exhibit sex differences with any electrode pairs (body fat:  $p = 0.09$ ; body water:  $p = 0.08$ ).

**Conclusion:** We conclude that sensitivity for perceiving electrical stimulation can be influenced by the subjects' characteristics, such as body fat or body water percentages.

**Keywords:** body fat percentage, current perception threshold, electrical stimulation, sex differences

**Conflict of Interest:** The authors have no conflict of interest to disclose; all experimental apparatuses were prepared using research funds from Kyorin University.

## INTRODUCTION

Quantitative sensory testing (QST) is a method for assessing peripheral nerve function, such as sensory threshold and estimating sensation magnitude and tolerance levels. Sensory threshold is the point at which sensation is consciously perceived. Determining the sensory threshold using QST has been employed by both researchers and clinicians, as it is a simple and painless test. Electrical stimulation is used for QST and has the advantage of controlling the magnitude of direct stimulation to intraepidermal nerve fibers.

Using a current perception threshold (CPT) measurement to determine sensory function is noninvasive, easy, and semi-objective; previous studies employing this technique have shown that sensitivity to electrical stimulation depends on several physiologic parameters, such as sex, age, physical sites, and presence of neuropathy (1–6). Although there are obvious differences in CPT between males and females, the reason for this difference remains unexplained. Past studies have suggested factors that affect sex differences in CPT, including the skinfold thickness or quantity of fat tissue (7–9). However, researchers have not confirmed whether these sex differences disappear after adjusting for the subjects' characteristics. If CPT data are influenced by body

characteristics, we assume that the sex differences in CPT can be suppressed using these parameters. To demonstrate our hypothesis, CPT is required to be clinically measured together with the subjects' characteristics. The analysis of their parameters is necessary to evaluate a more exact peripheral nerve function.

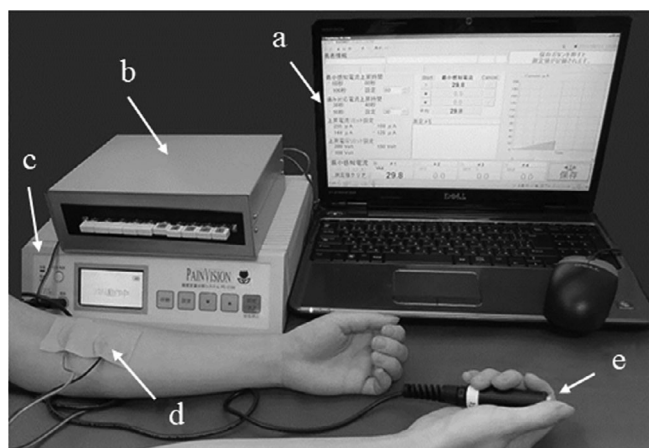
Address correspondence to: Shin-ichiro Seno, PhD, Faculty of Health Sciences, Kyorin University, 5-4-1 Shimorenjaku, Mitaka, Tokyo, Japan. Email: [seno@ks.kyorin-u.ac.jp](mailto:seno@ks.kyorin-u.ac.jp)

\* Faculty of Health Sciences, Kyorin University, Mitaka, Tokyo, Japan; and  
<sup>†</sup> Faculty of Health and Medical Sciences, Hokuriku University, Kanazawa, Ishikawa, Japan

For more information on author guidelines, an explanation of our peer review process, and conflict of interest informed consent policies, please go to <http://www.wiley.com/WileyCDA/Section/id-301854.html>

Source(s) of financial support: No external funding was available for this study, except for the support of Kyorin University.

This is an open access article under the terms of the Creative Commons Attribution-NonCommercial-NoDerivs License, which permits use and distribution in any medium, provided the original work is properly cited, the use is non-commercial and no modifications or adaptations are made.



**Figure 1.** Quantitative evaluation system for the current perception threshold (CPT) and pain magnitude: a. personal computer, b. electrode switch box, c. stimulator device (PainVision®), d. stimulating electrodes, and e. stop switch. A bipolar stimulating electrode is attached 6 cm from the medial cubital fossa of the left forearm. The distance between the proximal and distal electrodes is 1 cm in each case.

This study investigates the cause of sex differences in perception sensitivity for electrical stimulation and suggests an adjustment method that suppresses them. For this purpose, we measured the subject's characteristics (height, weight, muscle mass, body fat percentage, and body water percentage) and analyzed the relationship between them and CPT.

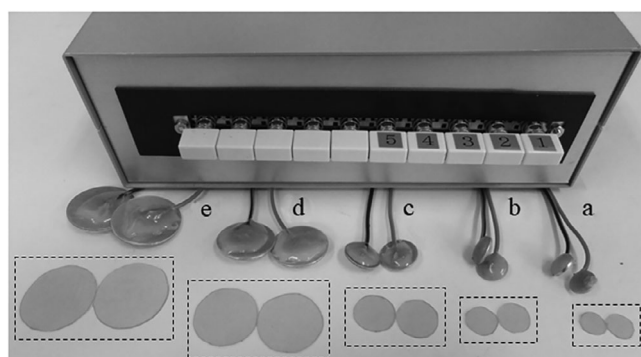
## MATERIALS AND METHODS

PainVision® (PS-2100, Nipro Corporation, Osaka, Japan) is a device that uses transcutaneous electrical stimulation to measure both CPT and the magnitude of pain sensation (Fig. 1) (1,10,11). This device is composed of an electrical stimulation system and a control system, which are driven by a built-in battery to protect subjects from the leakage current of an AC power supply.

The waveform of the stimulating current pulse was composed fundamentally of a square wave and was characterized by a sharp tip similar to a triangle wave; it repeated at 50 Hz (20 msec intervals) with a pulse duration of 0.3 msec. Using fast Fourier transform, the peak power was found at 50–2000 Hz. The rate of the waveform, at which the electrical current increased, was approximately 2.1  $\mu\text{A}/\text{s}$ . The mean current value of the electrical current in this system was recorded via a computer. We converted our mean current values to peak values by multiplying the mean current by pulse period: 20 msec/pulse duration: 0.3 msec because past CPT studies conventionally expressed their measurements as peak current values. We refer to the calculated peak current herein as the CPT.

## SUBJECTS

The subjects in this study included 70 healthy student volunteers (35 males and 35 females, aged 20–25 years). All subjects provided informed consent to participate in this study. The ethical committee of Kyorin University approved the study in advance under approval number 27-7.



**Figure 2.** Five types of bipolar stimulating electrodes and switch box. All electrodes are disk-shaped and 1.2 mm thick with a silver content of 92.5%. The diameter of each electrode is a. EL 1: 10 mm, b. EL 2: 12 mm, c. EL 3: 16 mm, d. EL 4: 25 mm, and e. EL 5: 30 mm. To maintain the equality of the electrical current distribution, a conductive gel sheet is attached between the skin and the electrodes for each subject. The conductive gel sheets are shown within the dashed lines in the figure.

## CUSTOMIZED ELECTRODES

To investigate sex differences in CPT, we prepared five sizes of bipolar-stimulating electrodes with silver disks that were 1.2 mm thick with a silver content of 92.5%. The diameter of electrodes EL 1, EL 2, EL 3, EL 4, and EL 5, respectively, was 10, 12, 16, 25, and 30 mm (as shown in Fig. 2). After brushing both sides of the silver disk with sandpaper, one side of each disk was connected to a lead wire using a conductive adhesive (Aremco-Bond 556; Audec Co., Ltd., Tokyo, Japan). The electrical resistance between the connecting wire and the silver disk was 10  $\Omega$  or less in all electrodes. Fresh electroconductive gel was attached directly to the other side of the electrode to maintain the equal distribution of the electrical current during measurement for each subject. The gel thickness was 0.6 mm. The electroconductive proportion of the lengthwise and lateral directions was approximately 50–100 times.

These stimulating electrodes were attached 6 cm from the medial part of the left cubital fossa for every measurement. We analyzed the data under the assumption that the location of the electrode did not vary with arm size.

## MEASUREMENT PROCEDURE

Basal information including identification number, sex, age, and body characteristics, such as height, weight, muscle mass, body fat percentage, and body water percentage were recorded before the CPT measurements. The characteristics other than height were measured with a body composition meter (BC-622, TANITA Corporation, Tokyo, Japan). After the stimulation site was sterilized using an alcohol-covered cotton swab and the stimulating electrode was attached, the amplitude of the current was automatically increased at approximately 2.1  $\mu\text{A}/\text{sec}$  until the subject pushed a stop switch upon first perceiving the electrical stimulation.

CPT was determined to be the mean of three measurements with each electrode. Additional measurements were required in the following cases: 1) the subject forgot to push the stop switch; 2) the subject requested a repeat measurement because they had pressed the switch too late; or 3) one of the three measurements differed by more than 20% from the other values. Throughout the experiment, we randomly attached the five sizes of electrodes to

**Table 1.** Comparison of the Physiologic Characteristics in the Healthy Subjects According to Sex.

	Males	Females	F-test	t-test
Number	35	35	–	–
Age	22.54 (±1.09)	22.37 (±0.91)	–	–
Height (cm)	170.65 (±5.53)	160.61 (±6.09)	$p = 0.65$ ( $F = 0.21$ )	$p < 0.001$ ( $t = 7.22$ )
Weight (kg)	63.88 (±9.99)	54.65 (±7.21)	$p = 0.17$ ( $F = 1.91$ )	$p < 0.001$ ( $t = 4.43$ )
BMI	21.89 (±2.96)	21.22 (±2.85)	$p = 0.93$ ( $F = 0.01$ )	$p = 0.33$ ( $t = 0.98$ )
Muscle mass (kg)	49.63 (±4.80)	36.49 (±3.01)	$p = 0.01$ ( $F = 6.41$ )	$p < 0.001$ ( $t = 13.71$ )
Body fat percentage (%)	17.25 (±5.87)	28.56 (±5.90)	$p = 0.74$ ( $F = 0.11$ )	$p < 0.001$ ( $t = -8.04$ )
Body water percentage (%)	54.26 (±8.31)	49.45 (±3.50)	$p = 0.01$ ( $F = 7.69$ )	$p = 0.003$ ( $t = 3.16$ )

each subject to overcome the influence of the order of attachment. A preliminary experiment was performed on all the subjects so that they were familiar with the sensation of electrical stimulation.

## STATISTICAL ANALYSIS

Statistically significant differences in the mean values of body characteristics between males and females were assessed with a Student's *t*-test if variances were equal as determined by an *F*-test and with Welch's *t*-test otherwise.

This study has performed the statistical analysis after having transformed the CPT into the log values. The Shapiro–Wilk's test was used to assess normal distribution.

The effect of the log-transformation values in all electrode sizes was adjusted for the body characteristics using an analysis of covariance (ANCOVA). Subsequently, the factors that influenced the CPT were identified using multiple regression analysis through a forcible loading method.

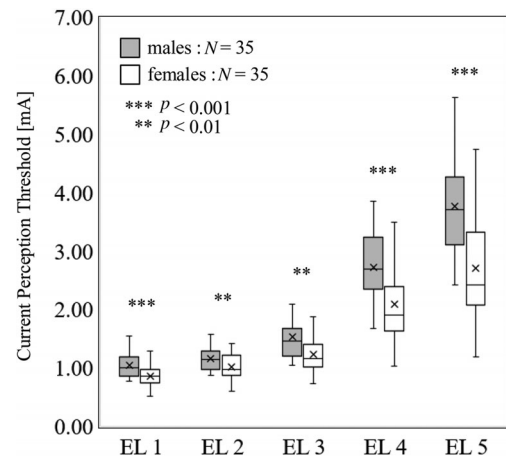
The homogeneity of the data variances was analyzed using the Levene's test. In the case of equal variances, a repeated-measures two-factor analysis of variance (ANOVA) was used to examine the differences in the log-transformation values between the sexes and the electrode sizes. If the analysis results violated the sphericity assumption, we applied Greenhouse–Geisser correction. In the ANOVA results, when significant main and interaction effects were detected, post hoc multiple comparisons were conducted by the Bonferroni method.

The software package SPSS Statistics version 22.0 for Windows (IBM Inc., Armonk, NY, USA) was used for all statistical analyses. *p* values less than 0.05 were considered statistically significant.

## RESULTS

As shown in Table 1, we observed sex differences in almost all physiologic parameters: height:  $t(68) = 7.22$ ,  $p < 0.001$ ; weight:  $t(68) = 4.43$ ,  $p < 0.001$ ; BMI:  $t(68) = 0.98$ ,  $p = 0.33$ ; muscle mass:  $t(57.14) = 13.71$ ,  $p < 0.001$ ; body fat percentage:  $t(68) = -8.04$ ,  $p < 0.001$ ; and body water percentage:  $t(45.66) = 3.16$ ,  $p = 0.003$ .

Fig. 3 shows the CPT values obtained from each electrode by sex. For electrodes EL 1–5, the males' measurement values were 1.05, 1.15, 1.54, 2.72, and 3.76 mA; the females' results were 0.86, 1.01,



**Figure 3.** Comparison of the current perception thresholds (CPTs) using five electrode types in male ( $N = 35$ ) and female ( $N = 35$ ) subjects. Data are shown for each electrode and expressed as box plots indicating male (gray) and female (white) subjects. The box limits are the 25th and 75th percentiles. The whiskers denote the minimum and maximum values. The crosses indicate the mean values. The horizontal line within each box shows the median. The asterisks denote a significant difference between the sexes (\*\*\*)  $p < 0.001$ , (\*\*)  $p < 0.01$ . A comparison of the CPT values within both the male and female groups shows statistically significant differences across the five electrodes ( $p < 0.001$ , not shown).

1.23, 2.09, and 2.71 mA. As results of having transformed these CPT into the log values, the males' log-transformation values were 0.011, 0.054, 0.171, 0.424, and 0.564; the females' results were  $-0.074$ ,  $-0.004$ , 0.076, 0.297, and 0.409. After having observed the normal distribution of log-transformation values, the Levene's test confirmed the homogeneity of the data variances (EL 1:  $F = 0.28$ ,  $p = 0.60$ ; EL 2:  $F = 0.03$ ,  $p = 0.86$ ; EL 3:  $F = 0.21$ ,  $p = 0.89$ ; EL 4:  $F = 2.78$ ,  $p = 0.10$ ; EL 5:  $F = 4.51$ ,  $p = 0.04$ ). Significant main effects of sex and electrode size were apparent in log-transformation values (sex:  $F_{1, 68} = 23.52$ ,  $p < 0.001$ ; electrode size:  $F_{2.55, 173.35} = 841.50$ ,  $p < 0.001$ ). These effects were superseded by a significant interaction between sex and electrode size ( $F_{2.55, 173.35} = 6.05$ ,  $p = 0.001$ ). Multiple comparisons showed that log-transformation values obtained from all electrodes were higher in males than in females (EL 1:  $F_{1, 68} = 16.50$ ,  $p < 0.001$ ; EL 2:  $F_{1, 68} = 8.03$ ,  $p = 0.006$ ; EL 3:  $F_{1, 68} = 13.57$ ,  $p < 0.001$ ; EL 4:  $F_{1, 68} = 19.92$ ,  $p < 0.001$ ; EL 5:  $F_{1, 68} = 26.66$ ,  $p < 0.001$ ).

Each physiologic parameter was analyzed by ANCOVA to identify the factors influencing the log-transformation values. As a result, six factors may affect the log-transformation values (i.e., sex:  $F = 82.51$ ,  $p < 0.001$ ; height:  $F = 25.79$ ,  $p < 0.001$ ; weight:  $F = 4.09$ ,  $p = 0.044$ ; body fat percentage:  $F = 75.78$ ,  $p < 0.001$ ; muscle mass:  $F = 49.74$ ,  $p < 0.001$ ; and body water percentage:  $F = 61.99$ ,  $p < 0.001$ ). Multiple regression analysis was performed with the forcible loading method, in which the dummy variables of the electrode (i.e., EL 1, EL 3, EL 4, and EL 5) and the variables (i.e., height, body fat percentage, and body water percentage) were significantly related to the log-transformation values. Three factors, namely sex, weight, and muscle mass, were excluded because of a promise of multicollinearity to other factors. The analysis results showed that the height ( $p = 0.022$ ), body fat percentage ( $p = 0.013$ ), body water percentage ( $p = 0.003$ ), and four electrode sizes (EL 1;  $p = 0.002$ , EL 3;  $p < 0.001$ , EL 4;  $p < 0.001$ , EL 5;  $p < 0.001$ ) had a significant effect on the log-transformation values (Table 2). Using these factors, a multiple linear regression equation is presented as follows:

$$Y = 0.002X_1 - 0.003X_2 + 0.004X_3 - 0.056EL1 + 0.098EL3 + 0.336EL4 + 0.461EL5 - 0.440$$

**Table 2.** Multiple Regression Analysis for the Log-Transformation Values (CPT Values) With Body Characteristics and Stimulating Electrodes.

Variable	<i>B</i>	<i>SE B</i>	$\beta$	<i>t</i> value	<i>p</i> -value
Height	0.002	0.001	0.067	2.308	0.022
Body fat percentage	-0.003	0.001	-0.105	-2.508	0.013
Body water percentage	0.004	0.001	0.111	2.953	0.003
Electrode (EL 1)	-0.056	0.018	-0.097	-3.106	0.002
Electrode (EL 3)	0.098	0.018	0.170	5.441	<0.001
Electrode (EL 4)	0.336	0.018	0.580	18.586	<0.001
Electrode (EL 5)	0.461	0.018	0.797	25.546	<0.001
Constant term	-0.440	0.197		-2.241	0.026
Adjusted $R^2$	0.787 ( $p < 0.001$ )				

The model predicts log-transformation values, *Y*, for healthy subjects with the perception sensitivity to the electrical stimulation: height,  $X_1$ ; body fat percentage,  $X_2$ ; body water percentage,  $X_3$ ; and number of electrodes, EL. This model had a coefficient of multiple determination,  $R^2$ , of 0.787 ( $p < 0.001$ ).

## CONCLUSIONS

We conclude that statistically significant differences in perception sensitivity for electrical stimulation can be induced by a subject's characteristics. Sex differences were observed in the obtained CPT values for all electrodes but could be suppressed by adjusting for body fat percentage or body water percentage. This report suggests a beneficial method for resolving sex differences in sensory sensitivity for electrical stimulation. In addition, this adjustment method could be extended for the subjects with suspected higher CPT (not abnormal peripheral nerve function).

## DISCUSSION

This study aimed to investigate the cause of sex differences in the perception threshold for electrical stimulation and provide an adjustment method to suppress these differences. The measurement values obtained from each electrode increased with the electrode size. Moreover, the CPT values measured for all electrodes were statistically significantly higher in males than in females. The multiple regression analysis suggested that the body fat percentage and the body water percentage are affectors for the log-transformation values.

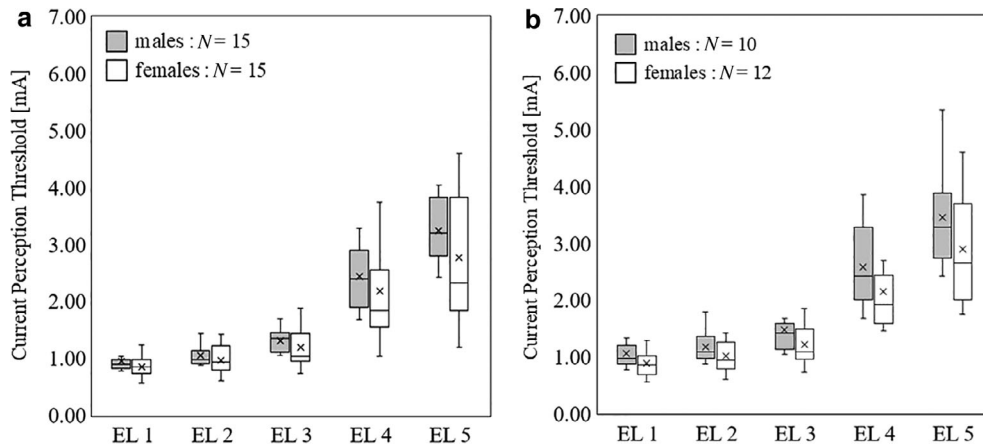
We analyzed whether the sex differences in the perception threshold are suppressed using two parameters. As shown in Table 1, two parameters in the participating subjects were used to observe the sex differences. Thus, we selectively chose more than 10 subjects to adjust the sex differences of these parameters. In the case of the body fat percentage, the selected subjects were 15 males with high percentage and 15 females with low percentage. In the case of the body water percentage, the selected subjects were 10 males and 12 females with body water percentages of 50–55%. By analyzing the log-transformation values in these groups, we investigated the possibility of suppressing the sex differences in the perception sensitivity.

Fig. 4a shows the CPT value results from 15 males with high body fat percentage (average: 22.45%) and 15 females with low body fat percentage (average: 23.19%). The average body fat percentages of the two groups were not statistically significantly different ( $t = -0.55$ ,  $p = 0.58$ ). The adjusted CPT values from

electrodes EL 1–5 in males were 0.94, 1.04, 1.31, 2.44, and 3.24 mA, respectively, whereas those in females were 0.85, 0.96, 1.19, 2.18, and 2.76 mA, respectively. After having transformed CPT values into the log values and observed the normal distribution of these data, the Levene's test confirmed the homogeneity of the data variances, except for EL 5 (EL 1:  $F = 0.79$ ,  $p = 0.38$ ; EL 2:  $F = 2.29$ ,  $p = 0.14$ ; EL 3:  $F = 4.25$ ,  $p = 0.05$ ; EL 4:  $F = 2.51$ ,  $p = 0.12$ ; EL 5:  $F = 13.58$ ,  $p = 0.001$ ). A significant main effect of the electrode size was also observed ( $F_{1,96, 54.75} = 369.06$ ,  $p < 0.001$ ). However, a main effect of sex was not significant ( $F_{1, 28} = 3.00$ ,  $p = 0.09$ ). These effects were not superseded by the significant interaction between the factors ( $F_{1,96, 54.75} = 1.03$ ,  $p = 0.36$ ). A multiple comparison of the log-transformation values in both males ( $F_{4, 112} = 203.73$ ,  $p < 0.001$ ) and females ( $F_{4, 112} = 166.37$ ,  $p < 0.001$ ) showed statistically significant differences among the five electrodes.

Fig. 4b shows the CPT results for subjects with body water percentages of 50–55% (10 males, 53.5%; 12 females: 51.9%). The average body water percentages in the two groups showed statistically significant differences ( $t = 2.30$ ,  $p = 0.032$ ). The adjusted CPT values from electrodes EL 1–5 in males were 1.06, 1.17, 1.48, 2.58, and 3.46 mA; the adjusted CPT values in females were 0.88, 1.01, 1.21, 2.15, and 2.89 mA. After having transformed CPT values into the log values and observed the normal distribution of these data, the Levene's test confirmed the homogeneity of the data variances (EL 1:  $F = 0.12$ ,  $p = 0.73$ ; EL 2:  $F = 1.59$ ,  $p = 0.22$ ; EL 3:  $F = 0.80$ ,  $p = 0.38$ ; EL 4:  $F = 0.07$ ,  $p = 0.79$ ; EL 5:  $F = 2.36$ ,  $p = 0.14$ ). A significant main effect of electrode size on log-transformation values was apparent ( $F_{2,12, 42.37} = 268.56$ ,  $p < 0.001$ ). However, the main effect of sex was not significant ( $F_{1, 20} = 3.38$ ,  $p = 0.08$ ). Moreover, these effects were superseded by no significant interaction between the factors ( $F_{2,12, 42.37} = 0.13$ ,  $p = 0.89$ ). Multiple comparisons of the log-transformation values showed statistically significant differences for each electrode both in males ( $F_{4, 80} = 126.90$ ,  $p < 0.001$ ) and in females ( $F_{4, 80} = 143.28$ ,  $p < 0.001$ ). These results suggested that the log-transformation values no longer exhibit a sex difference by adjusting for body fat or body water percentage.

Previous studies have reported sex differences in perception sensitivity for electrical stimulation regardless of electrode type or stimulation site (1,2,7–9,11). We observed similar results in this study for all electrodes. One earlier study demonstrated a link between subcutaneous adipose tissue mass and sensory current, suggesting that one affector of sensory current is skinfold thickness (7). Some researchers have suggested that the perception threshold for electrical stimulation may be related to body fat percentage (8). However, research had not confirmed whether sex differences disappear from measurement data after adjusting for those factors. Thus, we investigated the relationship between CPT and subjects' characteristics.



**Figure 4.** CPT comparison adjusting the body water percentage and body fat percentage. Data are shown for each electrode and expressed as box plots for males (gray) and females (white). The box limits are the 25th and 75th percentiles. The whiskers indicate the minimum and maximum values. The crosses denote the mean. The horizontal lines are the median values. a. depicts the data measured from 15 males and 15 females with similar body fat percentages (male average: 22.45%, female average: 23.19%,  $p = 0.58$ ). After having transformed CPT values into the log values, an ANOVA shows no significant main effect of sex for the transformation value ( $p = 0.09$ ), whereas a significant main effect of the electrode size is observed ( $p < 0.001$ ). Multiple comparisons of the log-transformation values in both males ( $p < 0.001$ ) and females ( $p < 0.001$ ) exhibit statistically significant differences for values among the five electrodes. b. shows an analysis of subjects with 50–55% body water percentage (10 males: 53.5%; 12 females: 51.9%,  $p = 0.032$ ). After having transformed CPT values into the log values, an ANOVA indicates that the main effect of the electrode size for the transformation value denotes a significant difference ( $p < 0.001$ ), whereas that of sex for the log-transformation value is not significant ( $p = 0.08$ ). Multiple comparisons of the CPT values show statistically significant differences for each electrode in both males ( $p < 0.001$ ) and females ( $p < 0.001$ ).

Although not shown in this article, the observed data from subject within normal limits for body fat percentage (males: 11–21%, females: 21–34%) exhibited significant sex differences, except in the case of EL 2. However, sex differences were not exhibited by analyzing data from males of high body fat percentage and females of low body fat percentage. These results support previous reports (7–9), and suggest that body fat percentage affects perception threshold. To explain the relationship between body fat percentage and perception threshold, we considered an anatomical tissue structure under the electrode. The skin comprises the epidermis, dermis, and subcutaneous tissue. The stimulating electric current flows toward a cathode from an anode through three layers. The epidermis or the dermis in healthy subjects would be slightly affected by the changes in body characteristics, whereas the thickness of the subcutaneous tissue in an obese subject markedly increased compared with that in a lean subject.

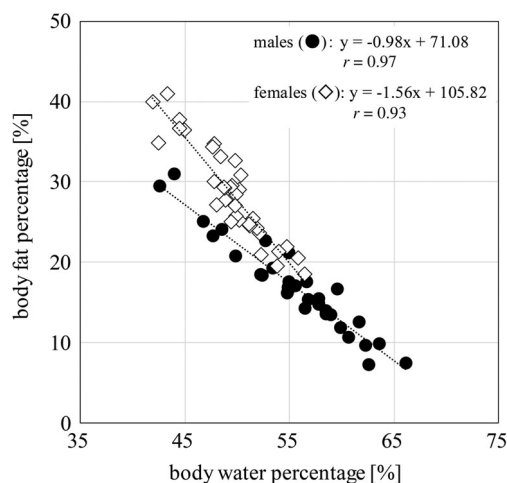
An electrical conductivity in the fat tissue is lower than that in the muscle or blood (12); hence, by the change of this tissue structure, most electrical currents will flow through the epidermis and the dermis and be converged in the region larger than the electrode area of the cathode when electrical stimulation is added to subjects with a high body fat percentage. As a result, epidermal nerve fibers are more easily excited as compared with those of subjects whose body fat percentage is low, resulting in a decreased perception threshold. In fact, CPT values in males were categorized into three groups by body fat percentage: low (less than 11%), normal (11–21%), and high (more than 21%). As shown in Table 3, these results validated our discussion and may suggest the possibility that a CPT determinant depends on the firing rate of the dermal nerve fibers among the anode and the cathode. However, a part of the results pertaining to female participants did not demonstrate a similar tendency, and these are not shown in this article. If the CPT values are determined by the summation of the electrical current under the cathode, the CPT could be measured by fully separating the anode and the cathode to test the validity of this discussion.

The CPT values within the normal limits of body water percentage (males: 55–65%, females: 45–60%) showed significant sex differences for all electrodes. However, these sex differences disappeared by adjusting for a body water percentage of 50–55%, likely because body water percentage and body fat percentage are highly correlated (13,14). In this study, Fig. 5 shows the relationship of two parameters divided by sex (35 males:  $r = 0.97$ , 35 females:  $r = 0.93$ ). Thus, it stands to reason that sex differences in CPT disappear after adjusting for body water percentage. Our method, adjusting to within 50–55% body water, is easier and may be suitable for analyzing sex differences in CPT.

We have considered that individual sensitivity to electrical stimulation, and sex differences of CPT, may be influenced by the number of epidermal nerve fibers. The density of intra-epidermal nerve fibers (IENFs) is decreased in elderly people (15,16) and

**Table 3.** Comparison of the CPT Values for the Three Categories of Male Participants.

	Body fat percentage		
	Under 11%	11–21%	More than 21%
Number	5	22	8
EL 1 (mA)	1.25 (±0.26)	1.02 (±0.22)	1.02 (±0.21)
EL 2 (mA)	1.30 (±0.24)	1.14 (±0.23)	1.09 (±0.19)
EL 3 (mA)	1.58 (±0.27)	1.60 (±0.54)	1.34 (±0.22)
EL 4 (mA)	3.07 (±0.26)	2.73 (±0.59)	2.46 (±0.51)
EL 5 (mA)	4.71 (±0.73)	3.71 (±0.87)	3.31 (±0.58)



**Figure 5.** Relationship between body water percentage and body fat percentage for all participants. The data are expressed as a scatter diagram comprising the aspects of both male (●;  $N = 35$ ) and female (◇;  $N = 35$ ) participants. The body water and body fat percentages (males:  $r = 0.97$ ; females:  $r = 0.93$ ) of the participants are strongly correlated.

patients with diabetes mellitus (17,18). Furthermore, CPT values in these groups are higher compared to those in healthy subjects (1,4–6). Similarly, females are reported to have more IENFs than males do (19,20). Thus, sex differences in CPT values could be explained by the distribution density of IENFs. In fact, differences in IENF quantity are posited to alter the thresholds of warmth and cold perception (21). Taken together, the number of IENFs under the stimulating electrode must play a key role in sensitivity to electrical stimulation.

We recognize that this study has a few limitations. First, all subjects were 70 healthy volunteer college students in their twenties from Kyorin University; results could potentially differ when a number of subjects of various groups are tested. Second, whether the change in the CPTs was concurrent with the change in the body fat percentage or body water percentage is unclear because this study did not perform a long-term follow-up of each subject. Third, whether the sex differences in the CPTs of neuropathy patients disappeared after adjusting the body fat percentage or body water percentage is unclear because the CPT increases were strongly affected by factors other than sex. In addition, the CPTs obtained using PainVision® were also likely under the influence of central sensitization in chronic pain conditions. Thus, the evaluation of the CPT in clinical fields is not only an evaluation of the peripheral nerve function but should also be considered with regard to this influence. To validate these results, it will be necessary to perform this experiment at many institutions, including subjects with a variety of characteristics.

## Authorship Statements

Shin-ichiro Seno planned the study concepts and design, conducted the experiments, and analyzed the data. Eiki Kogure, Atsushi Watanabe, and Hiroko Kobayashi contributed to the critical revision of manuscript. Hideaki Shimazu provided important intellectual input to complete this manuscript. All authors approved the final version of this manuscript.

## How to Cite this Article:

Seno S, Shimazu H, Kogure E, Watanabe A, Kobayashi H. 2019. Factors Affecting and Adjustments for Sex Differences in Current Perception Threshold With Transcutaneous Electrical Stimulation in Healthy Subjects. *Neuromodulation* 2019; 22: 573–579

## REFERENCES

- Seno S, Kato S, Kobayashi H, Watanabe A, Shimazu H. Evaluation of sex and age differences in the perception threshold of body surface against electrical stimulation: measurement of perception threshold to determine the possibility of diabetic neuropathy diagnoses. *Trans JSMBE* 2011;49:163–169. [in Japanese].
- Dalziel CF. Electric shock hazard. *IEEE Spectrum* 1972;9:41–50.
- Rollman GB, Harris G. The detectability, discriminability, and perceived magnitude of painful electrical shock. *Percept Psychophys* 1987;42:257–268.
- Takekuma K, Ando F, Niino N, Shimokata H. Age and gender differences in skin sensory threshold assessed by current perception in community-dwelling Japanese. *J Epidemiol* 2000;10:33–38.
- Matsutomo R, Takebayashi K, Aso Y. Assessment of peripheral neuropathy using measurement of the current perception threshold with the neurometer in patients with type 2 diabetes mellitus. *J Int Med Res* 2005;33:442–453.
- Masson EA, Veves A, Fernando D, Boulton AJ. Current perception thresholds: a new, quick, and reproducible method for the assessment of peripheral neuropathy in diabetes mellitus. *Diabetologia* 1989;32:724–728.
- Maffiuletti NA, Herrero AJ, Jubeau M, Impellizzeri FM, Bizzini M. Differences in electrical stimulation thresholds between men and women. *Ann Neurol* 2008;63: 507–512.
- Geng B, Yoshida K, Jensen W. Impacts of selected stimulation patterns on the perception threshold in electrocutaneous stimulation. *J Neuroeng Rehabil* 2011;8: 9. <https://doi.org/10.1186/1743-0003-8-9>.
- Leong GW, Lauschke J, Rutowski SB, Waite PM. Age, gender, and side differences of cutaneous electrical perceptual threshold testing in an able-bodied population. *J Spinal Cord Med* 2010;33:249–255.
- Shimazu H, Seno S, Kato S, Kobayashi H, Akimoto M. Development of a quantitative measurement method for the magnitude of pain using painless electrical stimulation and its evaluation using experimental pain. *Trans JSMBE* 2004;43: 117–123. [in Japanese].
- Seno S. Perception threshold by the electrical stimulation on oral cavity and lip regions. *Trans JSMBE* 2011;49:925–931. [in Japanese].
- Schwan HP. Electrical properties of tissue and cell suspensions. In: Lawrence JH, Tobias CA, editors. *Advances in biological and medical physics*. Volume 5. Cambridge, MA: Academic Press, 1957; p. 147–209.
- Fuller NJ, Jebb SA, Laskey MA, Coward WA, Elia M. Four-component model for the assessment of body composition in humans: comparison with alternative methods, and evaluation of the density and hydration of fat-free mass. *Clin Sci* 1992;82:687–693.
- Durnin JVGA, Womersley J. Body fat assessed from total body density and its estimation from skinfold thickness: measurements on 481 men and women aged from 16 to 72 years. *Br J Nutr* 1974;32:77–97.
- McArthur JC, Stocks EA, Hauer P, Cornblath DR, Griffin JW. Epidermal nerve fiber density: normative reference range and diagnostic efficiency. *Arch Neurol* 1998; 55:1513–1520.
- Kelly EJ, Terenghid G, Hazard A, Wiberg M. Nerve fibre and sensory end organ density in the epidermis and papillary dermis of the human hand. *Br J Plast Surg* 2005;58:774–779.
- Polydefkis M, Hauer P, Sheth S, Sirdofsky M, Griffin JW, McArthur JC. The time course of epidermal nerve fibre regeneration: studies in normal controls and in people with diabetes, with and without neuropathy. *Brain* 2004;127:1606–1615.
- Sorensen L, Molyneaux L, Yue DK. The relationship among pain, sensory loss, and small nerve fibers in diabetes. *Diabetes Care* 2006;29:883–887.
- Gøransson LG, Mellgren SI, Lindal S, Omdal R. The effect of age and gender on epidermal nerve fiber density. *Neurology* 2004;62:774–777.
- Lauria G, Bakkers M, Schmitz C et al. Intraepidermal nerve fiber density at the distal leg: a worldwide normative reference study. *J Peripher Nerv Syst* 2010;15: 202–207.
- Løseth S, Lindal S, Stalberg E, Mellgren I. Intraepidermal nerve fibre density, quantitative sensory testing and nerve conduction studies in a patient material with symptoms and signs of sensory polyneuropathy. *Eur J Neurol* 2006;13: 105–111.

**COMMENT**

Whenever one tries to evaluate and standardize the settings of transcutaneous electrical stimulation, the issues of threshold of perception and its variability become a topic of discussion. The authors of this study try to elucidate the potential reasons for such variability and link the body composition with gender differences. As mentioned by the authors, gender differences in perception sensitivity for electrical stimulation have been well described in the past on multiple occasions. However, the reasons for such differences remain

largely unknown. Although the paper does not answer all questions and provide all explanations, its findings may help future investigators to adjust expected threshold by considering the body fact composition and perhaps add it to the previously investigated skin fold thickness.

Konstantin Slavin, MD  
Chicago, IL, USA

Comments not included in the Early View version of this paper.