



# Effects of transcutaneous electrical nerve stimulation and visuotactile synchrony on the embodiment of an artificial hand

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## Abstract

The rubber hand illusion (RHI) is an experimental paradigm known to produce a bodily illusion. Transcutaneous electrical nerve stimulation (TENS) combined with the RHI induces a stronger illusion than the RHI alone. Visuotactile stimulus synchrony is an important aspect of the RHI. However, the effect of TENS and visuotactile stimulus synchrony in TENS combined with the RHI remains unknown. The purpose of this study was to investigate the effects of TENS and visuotactile stimulus synchrony on the embodiment of an artificial hand when using TENS combined with the RHI. The participants underwent four experimental conditions in random order: TENS/noTENS × Synchronous/Asynchronous. TENS was set at an intensity such that it generated a feeling of electrical paresthesia in the radial nerve area of the hand but did not cause pain, i.e., 100-Hz pulse frequency, 80- $\mu$ s pulse duration, and a constant pulse pattern. A visuotactile stimulus, either temporally synchronous or asynchronous, was generated using paintbrush strokes. To evaluate the outcome measures, the participants completed a questionnaire report and proprioceptive drift assessments (motor response and perceptual response). There were significant main effects of TENS and visuotactile synchrony, but no interaction between these factors, on the results of the questionnaire and the perceptual response. In contrast, there was no significant effect on the result of the motor response. These findings indicate that TENS and visuotactile synchrony might affect differently the embodiment of an artificial hand when using TENS combined with the RHI.

**Keywords** Rubber hand illusion · Transcutaneous electrical nerve stimulation · Visuotactile synchrony · Sense of body ownership · Proprioceptive drift

## Introduction

Embodiment is the sense of being localized within one's bodily borders (Arzy et al. 2006; Giummarra et al. 2008). Embodiment is not confined to the bodily self and may extend to a habitually used tool or prosthesis that effectively extends the body's area of influence. Moreover, embodiment is a complex multisensory process that involves representation of the bodily self (Giummarra et al. 2008). When we move our hands and feet, the sense that we are causing or

generating this action is the sense of agency, while the sense that the body we move belongs to us, and that we are mentally aware of it, is the sense of body ownership (Gallagher 2000). The sense of agency and the sense of body ownership reflect dissociable components of embodiment (Gallagher 2000; Longo et al. 2008). These sensations constitute a minimal sense of self (Gallagher 2000; Tsakiris et al. 2006). Gallese and Sinigaglia (2010) suggested that a minimal sense of self as the bodily self is necessary for and antecedent to both the sense of agency and the sense of ownership.

An illusion related to the sense of body ownership is the rubber hand illusion (RHI) (Botvinick and Cohen 1998; Tsakiris and Haggard 2005; Tsakiris et al. 2006). In the RHI, a participant's real hand, which is hidden from the participant's view, and an artificial rubber hand receive a tactile stimulus synchronously. This may cause the illusion that the artificial hand belongs to one's own body (Botvinick and Cohen 1998; Tsakiris and Haggard 2005). Botvinick and Cohen (1998) suggested that the RHI was evidence of the

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sensory information processing of three sensory modalities: touch, vision, and proprioception. Importantly, it is known that the RHI is greatly induced when tactile stimulation to an artificial hand and to the real hand are administered synchronously (Botvinick and Cohen 1998; Ehrsson et al. 2004; Shimada et al. 2009; Tsakiris et al. 2007). In the RHI, the sense of body ownership toward the artificial hand is measured through subjective ratings using a questionnaire (Botvinick and Cohen 1998; Ehrsson et al. 2004, 2005; Longo et al. 2008; Tsakiris and Haggard 2005). The attribution is measured quantitatively as a drift of the perceived position of one's own hand toward the artificial hand, using two types of measures pertaining to the proprioceptive drift: the motor response and the perceptual response (Kammers et al. 2009a, b; Riemer et al. 2013). Each measure is considered to reflect different body representations. The motor response concerns the body schema and the perceptual response concerns the body image (Kammers et al. 2009a, b; Riemer et al. 2013). The body schema is defined as an implicit reference frame for the guidance of movements, whereas the body image consists of conscious perceptions and attitudes toward one's own body (Gallagher 2005; Head and Holms 1911; Riemer et al. 2013). The body schema is an automatic, bottom-up sensory and organizational process; in contrast, the body image concerns higher-order, top-down bodily and perceptual representations (Giummarra et al. 2008; Kammers et al. 2009a). The distinction between the body schema and the body image is supported by clinical case studies (Gallagher 2011; Head and Holms 1911). Several neurological disorders involve bodily perception, body ownership, and self-embodiment (Giummarra et al. 2008). Phantom limbs after amputations, unilateral hemi-neglect, and somatoparaphrenia after stroke are considered disorders of bodily perception and body ownership (Giummarra et al. 2008). Use of the RHI paradigm could inform the contributions of the body schema to the feeling of limb ownership and motor representations in amputees (Funk et al. 2005; Giummarra et al. 2008). Research investigating the mechanisms behind illusion induction in the RHI is useful in elucidating the pathology of disorders of bodily perception and ownership and for developing neurorehabilitation methods (Christ and Reiner 2014; Giummarra et al. 2008).

Transcutaneous electrical nerve stimulation (TENS) is often used for pain relief (Milne et al. 2001; Nnoaham and Kumbang 2008) and in many rehabilitation settings (Lin et al. 2017; Zhu et al. 2017). A possible mechanism by which TENS relieves pain is described by the gate control theory (Melzack and Wall 1965). In this theory, it is posited that TENS activates low-threshold and large-diameter nerve fibers (e.g., A $\beta$  fibers) that are irrelevant to pain. This activation by TENS inhibits the transmission of nociceptive information at the synapses in the central nervous system (Johnson and Bjordal 2011; Sluka and Walsh 2003). A previous study

showed that the addition of somatosensory input by TENS to a tactile stimulus induced by paintbrush stroking resulted in stronger embodiment of the artificial hand than only tactile stimulation induced by paintbrush stroking (Mulvey et al. 2012). Mulvey et al. (2012, 2015) suggested that TENS may be acting as a method of somatosensory input, increasing tactile inputs and facilitating the embodiment of an artificial hand. These studies also suggest that TENS could be used as a method of somatosensory input to enhance the embodiment of prosthetics or affected limbs in neurorehabilitation (Mulvey et al. 2012, 2015). However, the effect of visuotactile synchrony on the embodiment of an artificial hand using both TENS and the RHI has not been investigated.

In the present study, we investigated the effects of TENS and visuotactile synchrony on the embodiment of an artificial hand combining TENS with the RHI. We hypothesized that, when applying TENS combined with RHI, synchronous visuotactile stimulation would induce greater embodiment of the artificial hand than asynchronous visuotactile stimulation.

## Methods

### Participants

Thirty-six healthy adults (10 men; age range 20–27 years) were recruited for the present study from Niigata University of Health and Welfare. This study was planned in accordance with the principles of the Declaration of Helsinki, the contents of the research were verbally explained to all participants, and consent was provided by all participants in writing. The study protocols were approved by the ethics committee of Niigata University of Health and Welfare (Number: 17923–171206). All participants were naive to the RHI. All participants were right-handed, as assessed by the revised Edinburgh Handedness Inventory (Oldfield 1971), translated in Japanese.

### Procedure

The participants sat in comfortable chairs, holding both arms in the prone position on a wooden board on a desk that was located in front of them. The right arm was placed 38 cm away from the midline of the body. An artificial right hand (Sato Giken, Kyoto, Japan) was placed parallel to and 15 cm left of the participant's right hand (measured from the index fingers). A wooden sight divider was placed between the participant's right hand and the artificial hand so that the participant could see only the artificial hand. The participant's left hand was placed 23 cm away from the midline of their body. The artificial hand and the participant's left

hand were placed at the same distance (23 cm) away from the midline.

Before data were collected, two TENS electrodes (square,  $5 \times 5$  cm) were placed over the superficial radial nerve of the participant's right arm. One electrode was placed 1 cm from the wrist joint and the other electrode was placed proximally 1.5 cm away from the proximal edge of the first electrode. TENS used ITO ESPURGE (Ito Co., Ltd, Tokyo, Japan) and was set to emit a constant current, a pulse frequency of 100 Hz, and a pulse width of 80  $\mu$ s. The intensity of TENS was determined by the intensity at which the participant felt it to be "strong but comfortable and not painful." This TENS intensity threshold was chosen based on previous studies (Mulvey et al. 2012, 2015).

The experiment consisted of four experimental conditions: TENS/noTENS combined with synchronous (Synch)/asynchronous (Asynch) visuotactile stimulation. The participants received these four conditions in a random order. Each condition consisted of a pre-measurement phase (approximately 2 min), an illusion-induction phase (2 min), a post-measurement phase (about 5 min), and a break for TENS washout (10 min). The experimental time totaled 90 min, including the time required to explain the experimental procedure to the participants. In this study, two investigators performed all experiments. The first investigator conducted the induction phase. The second investigator conducted the pre- and post-measurements and was blinded to the experimental conditions. In the illusion-induction phase in the TENS conditions, TENS was performed with a pre-set stimulus setting. In the illusion-induction phase of the noTENS conditions, TENS was not performed but the electrodes were still placed on the participant's right arm. In the illusion-induction phase in the synchronous conditions, the artificial hand and the participant's right hand were stroked synchronously. In the illusion-induction phase in the asynchronous conditions, the artificial hand and the participant's right hand were stroked asynchronously. The tactile stimuli were generated by two identical paintbrushes. Stroking was performed only on the area of the superficial radial nerve on both the artificial hand and the participant's hand. In the illusion-induction phase, the participant was instructed to watch the artificial hand being stroked while being unable to see their own right hand.

## Outcome measures

### Proprioceptive drift assessment

During the pre- and post-measurements, the participants were instructed to assess two types of proprioceptive drifts: the motor response and perceptual response. The participants were instructed to close their eyes. A wooden sight divider ( $20 \times 70 \times 8$  cm) was placed parallel to the wooden

board. The divider was placed to obstruct the participants' view of both their right hand and the artificial hand. After setting the divider, the participants first performed the motor response.

In the motor response, the participants were instructed to move their left index finger toward their right index finger in a single rapid ballistic movement. Specifically, the participants reached along the top of the wooden board and stopped the pointing movement at the position they considered identical to that of their right index finger. The pointing movements were always performed with eyes closed (Fig. 1a). The participants performed this motor response ten times for each measurement. The finger starting positions were randomly set from 11 to 18 cm from the midline of the participants' trunk. The second investigator guided the participants' left arm when returning to each starting position.

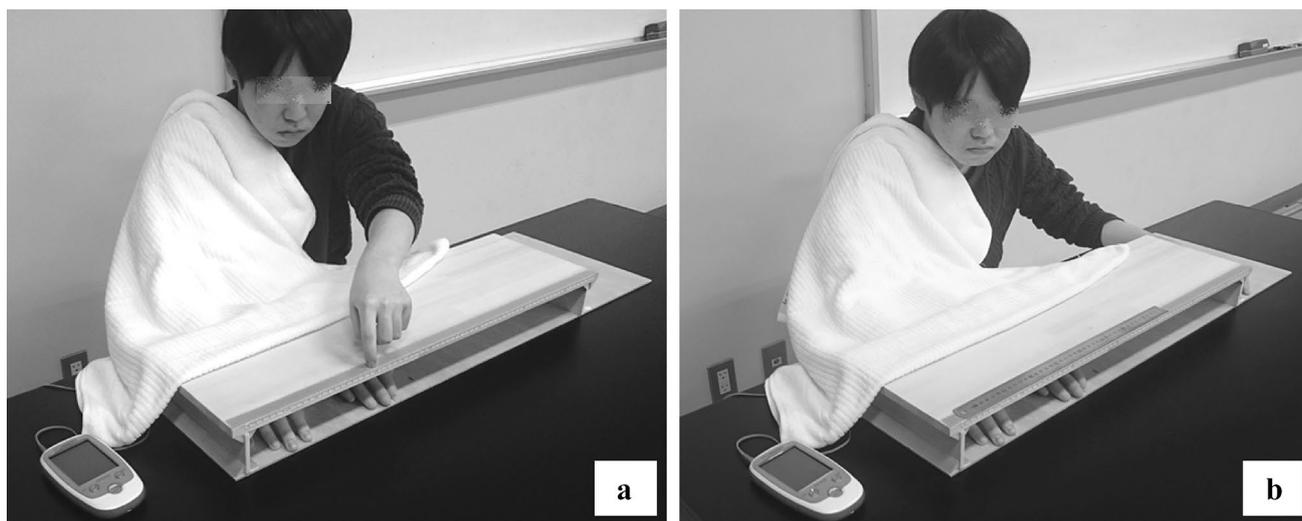
During the perceptual response, a ruler (40 cm) was set at the distal edge of the divider. After opening their eyes, the participants were instructed to verbally report the position of their right index fingertip based on the scale mark of the ruler (Fig. 1b). The position of the ruler was randomly moved horizontally 3 cm to the right or left from the participants' right index fingertip along the top of the wooden board for each response. The participants performed this perceptual response four times for each measurement. After measuring the perceptual responses of the proprioceptive drift, the participants were instructed to close their eyes again. Prior to the experiment, every participant practiced the required responses until each participant understood how to perform the two responses for measuring the proprioceptive drift.

### Questionnaire

After measuring the proprioceptive drift in the post-measurement phase of each condition, the participants answered a questionnaire to assess the subjective experience of the illusion. The questionnaire consisted of ten statements, presented in Table 1, derived from previous studies (Longo et al. 2008; Mulvey et al. 2012, 2015). Statements number 2, 3, and 9 were related to the sense of body ownership and the sense of agency (Mulvey et al. 2012, 2015). The participants answered these statements based on an 11-point Likert scale (0 = strongly disagree, 10 = strongly agree). This questionnaire and the answers given were in Japanese.

### Analysis

The motor-response and perceptual-response proprioceptive drift scores were calculated to measure differences between the pre-measurement and post-measurement scores, as well as the mean and standard deviation of the differences. A positive difference score indicated that the perceived position of



**Fig. 1** Setting of measurement of proprioceptive drift. **a** Motor response (left). The participant was instructed to point to the right index finger position with eyes closed. **b** Perceptual response (right).

The participant was instructed to report the position of the right index finger, as indicated by the scale mark, with eyes open

the participant's right hand drifted toward the artificial-hand side. The questionnaire scores were analyzed based on the mean scores of the statements relating to the sense of body ownership and the sense of agency (statements number 2, 3, and 9) and the scores of each statement (statements number 1 to 10). Before performing the statistical analysis, we examined the normality of the data using the Shapiro–Wilk test. When the data were normally distributed, we used a two-way (TENS factor: TENS/noTENS, stimulus-timing factor: Synch/Asynch) repeated measures ANOVA. When an interaction was identified, we used paired *t*-tests for post-hoc testing using Bonferroni correction. The partial  $\eta^2$  ( $\eta_p^2$ ) was calculated as the effect size. When the data were not normally distributed, we used Friedman's test. Kendall's *W* was calculated as the effect size. Statistical analysis was performed using R (version.3.4.1; R Foundation for Statistical Computing, Vienna, Austria).

## Results

### Proprioceptive drift assessment

Regarding the motor response, the mean  $\pm$  standard error (SE) of the difference between the pre- and post-measurements in each condition was as follows: noTENS/Asynch =  $1.57 \pm 0.47$ , TENS/Asynch =  $1.92 \pm 0.37$ , noTENS/Synch =  $1.61 \pm 0.42$ , and TENS/Synch =  $2.33 \pm 0.50$ . No significant effect was found using Friedman's test ( $\chi^2(3) = 2.17$ ,  $p = 0.538$ ,  $W = 0.020$ ) (Fig. 2a).

Regarding the perceptual response, the mean  $\pm$  SE of the difference between the pre- and post-measurements in each

condition was as follows: noTENS/Asynch =  $0.93 \pm 0.33$ , TENS/Asynch =  $1.95 \pm 0.39$ , noTENS/Synch =  $2.40 \pm 0.47$ , and TENS/Synch =  $2.95 \pm 0.54$ . There was a significant main effect of TENS ( $F_{(1,35)} = 6.30$ ,  $p = 0.017$ ,  $\eta_p^2 = 0.152$ ). There was also a significant main effect of stimulus timing ( $F_{(1,35)} = 12.48$ ,  $p = 0.001$ ,  $\eta_p^2 = 0.263$ ). There was, however, no interaction between TENS and stimulus timing ( $F_{(1,35)} = 0.58$ ,  $p = 0.453$ ,  $\eta_p^2 = 0.016$ ) (Fig. 2b).

### Questionnaire

When we analyzed the scores based on the questions regarding the sense of ownership and the sense of agency, the mean  $\pm$  SE of the score in each condition was as follows: noTENS/Asynch =  $3.64 \pm 0.42$ , TENS/Asynch =  $3.94 \pm 0.42$ , noTENS/Synch =  $6.31 \pm 0.32$ , and TENS/Synch =  $6.82 \pm 0.32$ . There was a significant main effect of TENS ( $F_{(1,35)} = 5.13$ ,  $p = 0.030$ ,  $\eta_p^2 = 0.653$ ). There was also a significant main effect of stimulus timing ( $F_{(1,35)} = 65.82$ ,  $p < 0.001$ ,  $\eta_p^2 = 0.128$ ). However, there was no interaction between TENS and stimulus timing ( $F_{(1,35)} = 0.96$ ,  $p = 0.333$ ,  $\eta_p^2 = 0.027$ ) (Fig. 3). The results of each statement in the questionnaire are shown in the Table. There was a significant interaction between TENS and the stimulus-timing factor only in the Q6 results. In the multiple comparisons, there was a significant difference between the noTENS/Synch (score  $2.47 \pm 0.41$ ) and noTENS/Asynch (score  $1.42 \pm 0.27$ ) ( $p = 0.031$ ) conditions, and there was no significant difference in the other comparisons ( $p > 0.05$ ).

**Table 1** Results of each statement of the questionnaire

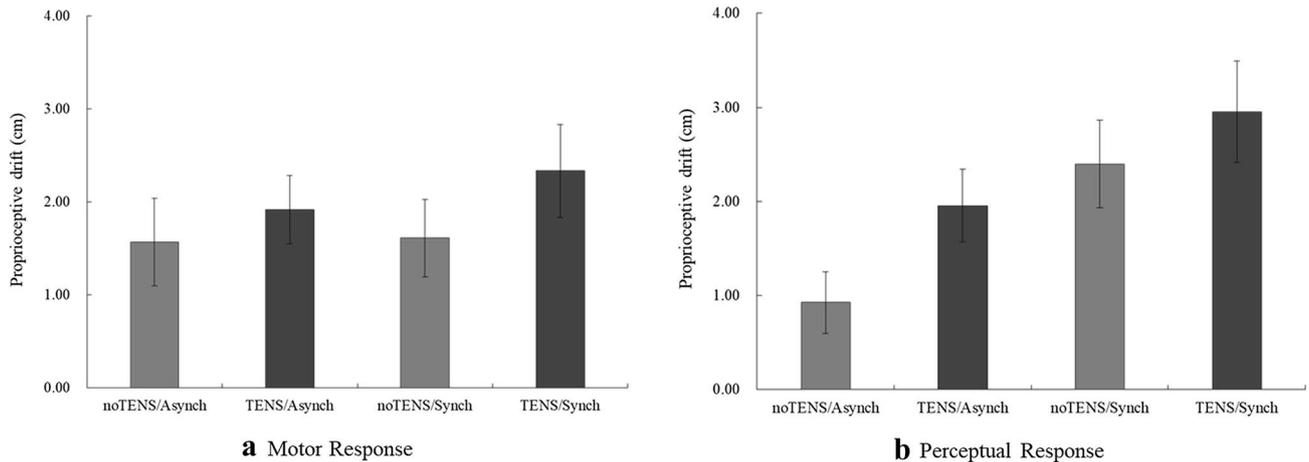
No.	Questionnaire statement	ANOVA factor	$F_{(1,35)}$	$p$	$\eta_p^2$
1	It seemed as if I were feeling the touch of the paintbrush in the location where I saw the artificial hand touched	TENS	0.87	0.357	0.722
		Stimulus timing	90.76	<0.001***	0.024
		TENS × stimulus timing	0.61	0.441	0.017
2	It seemed as if the artificial hand were my own	TENS	4.13	0.049*	0.106
		Stimulus timing	58.03	<0.001***	0.624
		TENS × stimulus timing	0.12	0.735	0.003
3	It seemed as if the artificial hand were part of my own body	TENS	1.33	0.257	0.037
		Stimulus timing	49.18	<0.001***	0.584
		TENS × stimulus timing	0.58	0.450	0.016
4	It seemed as if my real right hand were drifting towards the left	TENS	0.21	0.652	0.006
		Stimulus timing	14.76	<0.001***	0.297
		TENS × stimulus timing	0.39	0.538	0.011
5	It seemed as if I had two right hands	TENS	0.34	0.563	0.010
		Stimulus timing	0.16	0.695	0.004
		TENS × stimulus timing	2.45	0.127	0.065
6	It felt as if the artificial hand drifted slowly toward the right	TENS	0.17	0.681	0.005
		Stimulus timing	5.81	0.021*	0.142
		TENS × stimulus timing	4.92	0.033*	0.123
7	It felt as if the artificial hand and my own right hand lay closer to each other (compared to the beginning of the illusion-induction phase)	TENS	0.39	0.538	0.011
		Stimulus timing	10.00	0.003**	0.222
		TENS × stimulus timing	1.24	0.274	0.043
8	The artificial hand began to resemble my own hand in terms of shape, skin tone, freckles, or some other visual feature	TENS	0.09	0.765	0.003
		Stimulus timing	20.08	<0.001***	0.365
		TENS × stimulus timing	0.11	0.744	0.003
9	I felt as if when I moved my right hand, the artificial hand would move	TENS	4.60	0.039*	0.116
		Stimulus timing	34.77	<0.001***	0.498
		TENS × stimulus timing	0.65	0.424	0.018
10	My right hand felt numb	TENS	49.68	<0.001***	0.587
		Stimulus timing	0.27	0.605	0.008
		TENS × stimulus timing	0.09	0.762	0.003

\*\*\* $p < 0.001$ , \*\* $p < 0.01$ , \* $p < 0.05$

## Discussion

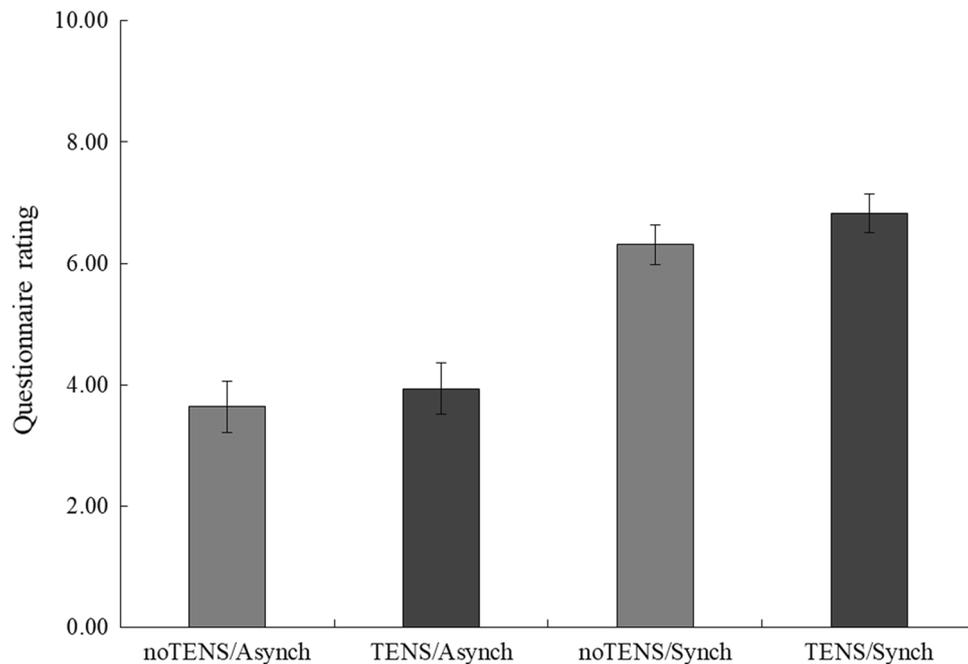
We investigated the effects of TENS and visuotactile stimulus synchrony on embodiment of an artificial hand using TENS combined with the RHI and questionnaire and proprioceptive drift assessments. We hypothesized that synchronous visuotactile stimulation would induce stronger embodiment of the artificial hand than asynchronous visuotactile stimulation when using TENS combined with the RHI, in line with the results of a previous study (Mulvey et al. 2012). However, there were no significant interactions between TENS and the stimulus-timing factor in the questionnaire and the perceptual proprioceptive drift results. These findings might indicate that TENS and visuotactile synchrony might affect differently the embodiment of the artificial hand when using TENS combined with the RHI.

In this study, there was a main effect of visuotactile synchrony on the questionnaire results and the perceptual proprioceptive drift. Previous studies have suggested a model of body ownership induction in the RHI (Makin et al. 2008; Tsakiris 2010). In this model, body ownership is induced following three comparisons of multisensory processing during the RHI. The first comparison is between the visual form of the artificial hand and the pre-existing body model. The second comparison is between the current state of the body and the postural and anatomical features of the body part that is to be experienced as one’s own. The third comparison is between the current sensory inputs, that is, between the seen and felt touches of the brush. Visuotactile synchrony provides positive feedback on this processing. That is, temporal synchrony between seen and felt events induces illusory binding of these events onto the visible artificial hand, which is experienced as the participants’ own (Costantini



**Fig. 2** Results of proprioceptive drift assessment. **a** Motor response (left). **b** Perceptual response (right). Gray bars indicate the noTENS conditions. Black bars indicate the TENS conditions. Error bars show standard errors across subjects. No significant effect was found using Friedman's test ( $\chi^2(3)=2.17$ ,  $p=0.538$ , Kendall's  $W=0.020$ ), (a). There were significant main effects of TENS ( $F_{(1,35)}=6.30$ ,

$p=0.017$ ,  $\eta_p^2=0.152$ ) and stimulus timing ( $F_{(1,35)}=12.48$ ,  $p=0.001$ ,  $\eta_p^2=0.263$ ) and no interaction between TENS and stimulus timing ( $F_{(1,35)}=0.58$ ,  $p=0.453$ ,  $\eta_p^2=0.016$ ) in the two-way ANOVA (b). Asynch: asynchrony, Synch: synchrony, TENS: transcutaneous electrical nerve stimulation



**Fig. 3** Results of the questionnaire questions related to body ownership and agency. The questionnaire rating scores presented correspond to the mean scores of three questionnaire statements (Q2, Q3, and Q9). These questionnaire statements are related to the bodily self. Gray bars indicate the noTENS conditions. Black bars indicate the TENS conditions. Error bars show standard errors across sub-

jects. Asynch: asynchrony, Synch: synchrony, TENS: transcutaneous electrical nerve stimulation. There were significant main effects of TENS ( $F_{(1,35)}=5.13$ ,  $p=0.030$ ,  $\eta_p^2=0.653$ ) and stimulus timing ( $F_{(1,35)}=65.82$ ,  $p<0.001$ ,  $\eta_p^2=0.128$ ), and no interaction between TENS and stimulus timing ( $F_{(1,35)}=0.96$ ,  $p=0.333$ ,  $\eta_p^2=0.027$ ) in the two-way ANOVA

et al. 2016; Makin et al. 2008). Conversely, visuotactile asynchrony provides negative feedback on this processing. Visuotactile stimulus asynchrony inhibits existing processes

of multisensory integration (Rohde et al. 2011). Some studies using the RHI have also shown that temporal synchrony of visuotactile stimuli is an important aspect for the

induction of the illusion (Botvinick and Cohen 1998; Ehrsson et al. 2004; Shimada et al. 2009; Tsakiris et al. 2007). Our results suggest that when we combine TENS with the RHI, visuotactile synchrony is an important aspect for the induction of bodily illusion.

This is consistent with the results of previous studies (Mulvey et al. 2012, 2015). These results suggest that TENS could be a novel method of somatosensory input for the embodiment of artificial limbs. Given that TENS activates low-threshold and large-diameter nerve fibers (e.g., A $\beta$  fibers) (Johnson and Bjordal 2011; Sluka and Walsh 2003), we hypothesized that when TENS is combined with the RHI, TENS would increase tactile input from A $\beta$  and similar fibers and facilitate embodiment of the artificial hand, with reference to the three-way weighted interaction between vision, touch, and proprioception for the RHI (Botvinick and Cohen 1998). However, we did not find an interaction between TENS and the stimulus-timing factor in this study. This indicates that, even if we combine TENS, as a method of tactile stimulation, with the RHI, TENS does not increase tactile input by paintbrush stroking. Hence, our results indicate that TENS affects multisensory processing in the embodiment of an artificial hand. Conversely, the effects of TENS might differ from the effects of visuotactile stimulation induced by paintbrush stroking. The results of Q1 and Q10 in the questionnaire indicate that participants regarded tactile inputs by a paintbrush and electrical paresthesia by TENS as different sensations. Mulvey et al. (2012, 2015) also discussed the difference in the somatosensory sensation derived by TENS and a paintbrush. Some studies have shown that pleasant tactile stimulation is important for induction of the RHI (Lloyd et al. 2013; van Stralen et al. 2014). They discussed that pleasant tactile stimulation, e.g., stroking at a low velocity on hairy skin using soft materials activated slow-conducting, unmyelinated, low-threshold mechano-receptive fibers (C tactile fibers) that project to the posterior insular cortex (Olausson et al. 2010) and modulate the RHI (Lloyd et al. 2013; van Stralen et al. 2014). The result of Q10 suggests that TENS caused electrical paresthesia, which was afferent from low-threshold and large-diameter nerve fibers (e.g., A $\beta$  fibers), not pleasant touch. In addition, in terms of the temporal pattern of somatosensory input, there might be a difference between the sensation derived from TENS and the paintbrush. TENS was delivered at a constant pattern of somatosensory input that was set at 100 Hz in this study. In contrast, tactile input by the paintbrush was delivered at a non-constant pattern of somatosensory input, administered by the experimenter. This experimental setting indicates that TENS was providing somatosensory input while the participant's real hand and the artificial hand were stroked or not in both the synchronous and asynchronous conditions. Therefore, we propose that the somatosensory input induced by TENS and the tactile input induced by paintbrush

stroking might be processed differently in the embodiment of an artificial hand.

We need to carefully consider the effect of TENS on embodiment of the artificial hand in the RHI. The result of Q10 indicates that TENS caused strong afferent alteration, perceived as numbness and not as pain. Some previous studies have reported that inducing anesthesia caused change in body representation. When participants underwent anesthesia using nerve block or ischemic pressure, they experienced change in body image (Proske and Gandevia 2012). Inducing anesthesia using nerve block caused loss of sensory input from small-diameter afferent fibers; it caused perceptual distortions of the body shape and body size (Gandevia and Phegan 1999; Paqueron et al. 2003). Inducing anesthesia using ischemic pressure caused loss of sensory input from large-diameter afferent fibers; it caused perceptual distortion of the body size and limb position (Inui et al. 2011; Walsh et al. 2015). The results of these studies suggest that alteration of afferent input might change embodiment of one's own body. Nevertheless, in our study, the short induction periods and low intensity, continuous afferent input by TENS might have altered the body image (perceptual proprioceptive drift) and the questionnaire results related to the sense of body ownership and the sense of agency. In contrast, there is the possibility that TENS would affect a different aspect of bodily self-perceptual alteration. The spatial-attention bias induced by the afferent input of TENS might influence the alteration of the bodily self. Previous studies have reported a relationship between spatial-attention bias and the RHI (Ocklenburg et al. 2012; Zeller and Hullin 2018). In the present study, participants felt a strong sensory afferent input that was not painful, as indicated from the result of Q10. Due to the afferent inputs by TENS, participants might actively direct spatial attention toward the artificial-hand space. In addition, we should consider the result of Q6 ("It felt as if the artificial hand drifted slowly toward the right"). There was an interaction between TENS and stimulus timing only in the Q6 results. In the Q6 results, there was a main effect of the stimulus timing, not the TENS, factor. This result might reflect a combined effect when we use TENS with the RHI. However, this result should be interpreted with caution because, it was inconsistent with the results of previous studies (Botvinick and Cohen 1998; Kammers et al. 2009a; Riemer et al. 2013; Rohde et al. 2011). In Q6, the agreement score of the noTENS/Synch condition was larger than that of the noTENS/Asynch condition. However, the scores in both conditions were low. In our study, there were significant effects of the stimulus synchrony factor in the control statements (e.g., Q4, 6, 7, and 8), which is inconsistent with the results of previous studies (Botvinick and Cohen 1998; Kammers et al. 2009a;

Riemer et al. 2013; Rohde et al. 2011). For these reasons, it is not possible to draw definitive conclusions regarding the combined effect on the subjective experience between TENS and stimulus synchrony, especially regarding Q6.

In this study, there was no significant effect of the experimental conditions on the motor response of the proprioceptive drift. Previous studies using the RHI have suggested that a questionnaire is able to assess the experience of the bodily self especially as it relates to body ownership, the perceptual response of the proprioceptive drift (assessing body image), and the motor response of the proprioceptive drift (assessing body schema) (Kammers et al. 2009a, b). These studies have suggested that questionnaire-related body ownership and proprioceptive drift, such as those used in our study, are different outcome measures and that they reflect different processing-related body representations (Kammers et al. 2009a, b; Rohde et al. 2011). Kammers et al. (2009a, b) reported that the perceptual response, but not the motor response, was susceptible to the bodily illusion. In our study, there was no significant effect of the experimental conditions on the body schema, as was the case in these studies (Kammers et al. 2009a, b). However, numerous previous studies (Botvinick and Cohen 1998; Fuchs et al. 2016; Kalckert and Ehrsson 2012; Riemer et al. 2013) have reported results that are inconsistent with our results regarding the motor response. In our study, participants were instructed to repeat the motor response task 10 times with random starting positions to avoid a learning effect regarding the hand's spatial position, in line with a previous study (Fuch et al. 2016). However, in the motor response, proprioceptive signals in the ballistic reach movement of the response hand might cause calibration of the participant's body schema and decrease the drift of the perceptual real hand position toward the artificial hand. During the assessment of the motor response, the after effect of alteration of the body schema might be reduced because of movement and the measurement time for the 10 motor responses. Hence, caution is needed when interpreting the results regarding the motor response in this study.

Our study has some limitations. In this study, it is not clear why or how TENS and visuotactile stimulus synchrony had differing effects on the induction of the RHI. Thus, future studies should investigate the effects of TENS on the sense of body ownership and/or limb position sense. Moreover, neuroimaging and neurophysiological research methods might be necessary to clarify the induction mechanisms underlying the effects of TENS and visuotactile synchrony in the RHI. Finally, in the current study, we included only healthy volunteers, limiting its clinical applicability. Regardless, these findings contribute to the future development of neurorehabilitation methods.

## Conclusion

In this study, we investigated the effects of TENS and visuotactile synchrony on the embodiment of an artificial hand using TENS combined with the RHI. The results of this study showed that TENS and visuotactile synchrony might affect differently the embodiment of an artificial hand. These results might provide us with novel perspective on the neurorehabilitation of diseases of body ownership and body perception.

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**Author contributions** AA and KS conceived of the study and designed the experimental paradigm. AA, KY, and YK performed the experiment and analyzed the data. AA wrote the manuscript. KS provided feedback and edited the manuscript. All authors read and approved the final manuscript.

## Compliance with ethical standards

**Conflict of interest** The authors declare that they have no conflict of interest.

**Ethical approval** All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards.

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