

ORIGINAL**Prefrontal activation during two Japanese Stroop tasks revealed with multi-channel near-infrared spectroscopy**

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Abstract : The Stroop task is sometimes used in psychiatric research to elicit prefrontal activity, which presumably reflects cognitive functioning. Although there are two Stroop tasks (Kana script and Kanji script) in Japan, it is unclear whether these tasks elicit the same hemoglobin changes. Moreover, it is unclear whether psychological conditions or characteristics influence hemoglobin changes in the Japanese Stroop task. The aim of this study was to clarify whether hemoglobin changes elicited by the two Japanese Stroop tasks accurately reflected cognitive functioning. Hemoglobin changes were measured with multi-channel near-infrared spectroscopy (NIRS) in 100 healthy Japanese participants performing two Japanese Stroop tasks. The Beck-Depression Inventory (BDI), State-Trait-Anxiety Inventory (STAI), and Maudsley Obsessive Compulsive Inventory (MOCI) were administered to participants to identify psychological conditions or personality characteristics. Compared with the Kanji task, the Kana task produced a greater Stroop effect and a larger increase in oxyhemoglobin (oxy-Hb) concentration. Moreover there were no significant correlations between oxy-Hb concentration and BDI, STAI-trait, STAI-state, or MOCI scores. Therefore we found that a participant's psychological conditions or characteristics did not influence the hemodynamic changes during either task. These data suggest the Kana Stroop task is more useful than the Kanji Stroop task for NIRS studies in psychiatric research. *J. Med. Invest.* 62 : 51-55, February, 2015

Keywords : Japanese Stroop tasks, near-infrared spectroscopy, prefrontal activity, psychological condition

INTRODUCTION

Near-infrared spectroscopy (NIRS) is a functional neuroimaging tool that can measure hemodynamic changes in the cerebral cortex. Multi-channel NIRS has been increasingly used in psychiatric research because it is a noninvasive, portable, and inexpensive technique compared with other functional neuroimaging tools, such as functional magnetic resonance imaging (fMRI), positron emission tomography (PET), and single-photon emission computed tomography (SPECT). When measuring changes in brain activity related to a specific cognitive function with NIRS, the subject performs a cognitive task related to the function that should modulate cortical activity, and subsequently, hemoglobin levels. NIRS has an advantage in that it allows subjects to perform cognitive tasks under relatively natural conditions when compared with those performed in an MRI scanner. The verbal fluency task (VFT) has been the most popular cognitive task performed in NIRS studies, whereas the Stroop task is sometimes used in psychiatric research to study prefrontal cerebral blood flow changes reflecting cognitive functions.

The Stroop task is a cognitive interference task in which the subject is shown a word written in colored letters, and in response, the subject names the color. The cognitive interference is introduced when the word is presented in a color that differs from the color the word represents semantically. When the letter color disagrees with the meaning of the word (incongruent condition), the subject will typically respond with a delayed discrimination time

and an increased in the number of false reactions compared to when the two attributes are consistent (congruent condition). This phenomenon is known as the Stroop effect or Stroop interference (1). As the Stroop task requires the ability to inhibit a habitual automatic response, it is widely used to assess executive functions.

The Stroop task has been used with neuroimaging techniques including fMRI, PET, and NIRS. Previous neuroimaging studies have reported the prefrontal cortex (PFC) is strongly activated during the Stroop task (2-4). fMRI studies have shown that the degree of the Stroop effect is strongly correlated with activation of the anterior cingulate cortex or inferior frontal gyrus bilaterally (5). NIRS studies have reported the dorsolateral PFC (DLPFC) and the inferior frontal gyrus are also activated by the Stroop task (6).

In Japan, there are two Stroop tasks because the Japanese language uses two orthographic systems, phonographic Kana and ideogrammatic Kanji, which are used in combination. Several studies have indicated a difference in the Stroop effect between the phonographic and ideogrammatic systems (7-9). Some neuroimaging studies have shown that different brain areas can be activated with Kana and Kanji (10-12). However, it is uncertain whether the Kana and Kanji Stroop tests elicit hemodynamic changes in the prefrontal cortex with similar amplitudes. Moreover, it is not clear whether certain psychological conditions or characteristics, such as depression, anxiety, or obsession, influence the hemodynamic changes elicited with the Japanese Stroop task. The purpose of this study was to clarify whether hemoglobin changes elicited by the two Japanese Stroop tasks accurately reflected cognitive functions, and which Stroop task may be more useful in psychiatric research.

Received for publication June 26, 2014 ; accepted September 9, 2014.

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METHODS

Participants

One hundred healthy, right-handed Japanese volunteers (50 men, 50 women) participated in the study. The participants' mean age was 24.7 years (range=21-40, SD=3.6 years). They had no history of major mental illness, neurological illness, traumatic brain injury, or substance abuse (individual or family). Written informed consent was obtained from each subject before they participated in this investigation; the Ethical Committee of the University of Tokushima approved the study. All participants were required to have normal color vision and normal linguistic-cognitive development. All participants were native Japanese speakers. The Beck-Depression Inventory (BDI) (13), State-Trait-Anxiety Inventory (STAI) (14), and Maudsley Obsessive Compulsive Inventory (MOCI) (15) were administered to measure the participants' tendency for depression, anxiety, and obsession, respectively.

Procedure

Each participant sat in a comfortable chair facing a computer screen. The computer screen displayed the words "red," "green," "yellow," or "blue" written in either Kana or Kanji script and printed in red, green, yellow, or blue. In the congruent condition, the word's semantic meaning was congruent with the script color (e.g., the word "green" was printed in green). In the incongruent condition, the words were printed in a color that was incongruent to the semantic meaning. For each task, 100 words were presented at once. Subjects were instructed to name as quickly as possible the color in which the words were printed. The investigator recorded the number of correct and incorrect responses.

The task was presented in a block design that included rest periods and four test conditions (congruent and incongruent conditions in both Kana and Kanji scripts). The four task conditions were separated by a rest period. In the rest periods, subjects were instructed to look at a dot presented on the computer screen. The duration of each task condition and rest period was 30 s. NIRS measurements were performed throughout the task, including the rest and all four task conditions. The task was administered in the following sequence: rest, Kana (or Kanji) congruent condition, rest, Kana (or Kanji) incongruent condition, rest, Kanji (or Kana) congruent condition, rest, and Kanji (or Kana) incongruent condition. To reduce the influence of an order effect, the Kana and Kanji tasks were presented in a counterbalanced order for fifty of the participants.

NIRS measurement

NIRS measurements were performed using an ETG4000 NIRS system with 24 optodes (Hitachi Medical Corporation, Tokyo, Japan), and using two wavelengths of near-infrared light (695 and 830 nm). The light absorption was measured and changes in oxyHb and deoxyHb concentrations were calculated according to the Beer-Lambert law using the difference in absorption between the two wavelengths. The distance between the emitter and detector was 3.0 cm; the NIRS measurements were taken from points located 2-3 cm beneath the scalp, which is the surface of cerebral cortex. The NIRS probes were placed bilaterally and symmetrically on the head. The 2 plastic shells with 9 optodes attached were placed over the frontal region, with one shell placed on the left side of the forehead and the other placed on the right side of the forehead. Each probe measured the relative changes in oxyHb and deoxyHb concentrations at 12 points within a 6 × 6 cm area over the respective hemisphere. The most inferiorly positioned probes were along the Fp1-Fp2 line in accordance with the International 10/20 Electrode Placement System for electroencephalography. The distances from the midline to the most medial and lateral probes were 1.5 cm and 7.5 cm, respectively. Data was

obtained and analyzed using the *Integral mode*. The pre-task baseline was the mean signal during the 10-s period immediately before the task period, and the post-task baseline was the mean signal during the 5-15 s immediately following the task period.

Data analysis and statistics

First, the number of correct responses was analyzed for the four task conditions (Kana congruent, Kana incongruent, Kanji congruent and Kanji incongruent) using a one-way analysis of variance (ANOVA). For both Kana and Kanji, the difference in performance between the two conditions (congruent-incongruent) was calculated as an index of the Stroop effect. The difference in the index between Kana and Kanji was examined by a paired *t*-test.

The levels of oxy-Hb, deoxy-Hb, and total-Hb concentration time-locked to the task were obtained. In order to increase the signal-to-noise ratio, data from the 24 channels were filtered with a digital band pass (0.0005-0.02 Hz). Our analysis focused on oxy-Hb changes because recent articles have reported that oxy-Hb is the most sensitive hemodynamic indicator of the brain's activation (16). To avoid Type I errors, data from the 24 channels recorded during each task condition were divided into four areas (Area 1 channels: 1, 2, 3, 4, 6, and 8; Area 2 channels: 5, 7, 9, 10, 11, and 12; Area 3 channels: 13, 14, 16, 17, 19, and 22; and Area 4 channels: 15, 18, 20, 21, 23, and 24) before the analysis (Figure. 1a).

The oxy-Hb concentration was averaged for each task condition (Kana congruent, Kana incongruent, Kanji congruent, and Kanji incongruent). For the statistical analysis, a one-way ANOVA was performed with oxy-Hb as the dependent variable. Next, the average peak change in oxy-Hb concentration in each area (Areas 1-4) was calculated for each participant. Data indicating changes in oxy-Hb concentrations were analyzed with a two-way repeated measures ANOVA using the variables (i) "orthographic systems" (Kana and Kanji) and (ii) "area" (Areas 1-4). Bonferroni's method was used as a post-hoc test. When necessary, Greenhouse-Geisser correction was applied to the degrees of freedom.

Person's correlation coefficient (*r*) was used to study the relationship between the oxy-Hb concentration and BDI, STAI-trait, STAI-state, and MOCI scores. Statistical significance was adjusted for multiple comparisons (Bonferroni's correction).

In the present analysis, the alpha level was set at 0.05; all statistical analyses were performed with SPSS.

RESULTS

Performance data

The results are summarized in Table 1. There was a main effect among the four conditions for the number of responses ($F(1,99)=5242.415, p<.001$). A post hoc *t*-test showed that the congruent condition had a greater number of correct responses than the incongruent condition for both Kana and Kanji (Kana: congruent > incongruent, $t=28.45, p<.001$; Kanji: congruent > incongruent, $t=18.75, p<.001$). However, the Kana congruent condition had more correct responses than the Kanji congruent condition (congruent: Kana > Kanji, $t=10.460, p<.001$); there was no significant difference in the number of correct responses between the Kana and Kanji incongruent conditions. For the number of errors, there was a main effect among the four conditions ($F(1,99)=126.108, p<.001$). Post hoc *t*-tests showed that both Kana and Kanji had significantly more errors in the incongruent condition than in the congruent condition (Kana: incongruent > congruent, $t=1.370, p<.001$; Kanji: incongruent > congruent, $t=0.710, p<.001$). For the congruent condition, Kanji had more errors than Kana (congruent: Kanji > Kana, $t=0.24, p<.01$). In contrast, there were no significant difference between Kana and Kanji in the incongruent

Table 1. Performance data (response and error)

	response		t-value	error		t-value	
	congruent	incongruent		congruent	incongruent		
Kana	69.44 ± 11.95	40.99 ± 7.59	28.450***	Kana	0.09 ± 0.35	1.46 ± 1.68	1.370***
Kanji	58.98 ± 10.42	40.23 ± 15.60	18.750***	Kanji	0.33 ± 0.68	1.04 ± 1.19	0.710***
t-value	10.460***	0.760 n.s.		t-value	0.24*	0.420 n.s.	

Number of response and error in kana and kanji task were presented. The significance level are shown with asterisks (* $p < .01$, *** $p < .001$, n.s. no significant).

condition. The Stroop effect in the performance data was compared between Kana and Kanji. The Stroop effect was significantly larger with Kana than with Kanji (response : Kana > Kanji, $t = 7.178$, $p < .001$; Error : Kana > Kanji, $t = 3.323$, $p < .01$).

NIRS data

In most channels, we observed the oxy-Hb signal increased during the Stroop test compared to baseline. The results of the one-way ANOVA for this analysis are presented in Table 2. We identified a significant main effect for task condition ($F(3, 297) = 11.758$, $p < .001$), and the post hoc test showed a significantly greater activation for the Kana incongruent condition than for the Kana congruent condition ($t = 0.058$, $p < .001$). In addition, the Kana incongruent condition showed a significantly greater activation than the Kanji incongruent condition ($t = 0.032$, $p < .05$). However, there were no significant differences between the Kanji incongruent and Kanji congruent conditions or between the Kana congruent and Kanji congruent conditions.

The difference in oxy-Hb concentration between the two conditions (incongruent-congruent) in both Kana and Kanji was calculated as an index that showed the level of the Stroop effect between the orthographic systems (Kana and Kanji). We conducted a two-way repeated measures ANOVA on the oxy-Hb levels using the two factors (orthographic systems ; Kana and Kanji) × (area ; Areas 1-4) ; the results are presented in Table 3. As for oxy-Hb,

there was a main effect for the orthographic system ($F(1, 99) = 4.410$, $p < .05$). The oxy-Hb levels for the Kana condition were significantly larger than those for Kanji condition ($t = 0.031$, $p < .05$). A main effect of area also was significant ($F(3, 297) = 14.282$, $p < .001$). Area 1 and Area 3 showed significantly greater activation than Area 2 and Area 4 (Area 1 > Area 2 ; $t = 0.053$, $p < .001$, Area 1 > Area 4 ; $t = 0.051$, $p < .001$, Area 3 > Area 2 ; $t = 0.060$, $p < .001$, Area 3 > Area 4 ; $t = 0.057$, $p < .01$). We identified a significant interaction between the orthographic system and brain area ($F(3, 297) = 3.153$, $p < .05$). Post hoc paired comparisons revealed that with Kana, oxy-Hb levels for Area 1 and Area 3 were larger than those for Area 2 and Area 4 (Area 1 > Area 2 : $t = 0.069$, $p < .001$; Area 1 > Area 4 : $t = 0.062$, $p < .01$; Area 3 > Area 2 : $t = 0.092$, $p < .001$; Area 3 > Area 4 : $t = 0.085$, $p < .01$). No significant differences were observed with Kanji (Figure. 1b).

Pearson’s correlations between oxy-Hb concentration and BDI, STAI-trait, STAI-state, and MOCI scores are presented in Table 4. After Bonferroni’s correction, there were no significant correlations between oxy-Hb concentration and BDI, STAI-trait, STAI-state, or MOCI scores. We also examined the correlation between performance (response and error) in the two Stroop tasks and BDI, STAI-trait, STAI-state, and MOCI scores. However, there was no significant correlation observed between performance and any scores.

Table 2. Oxy-Hb changes during Japanese Stroop test

task	condition	[oxy-Hb] (mmol · mm)	t-value				ANOVA	F-value
			Kana		Kanji			
			congruent	incongruent	congruent	incongruent	task condition	
Kana	congruent	0.175 ± 0.11	-	0.058***	0.001 n.s.	0.025 n.s.	11.758***	
	incongruent	0.232 ± 0.11	-	-	0.011***	0.032*		
Kanji	congruent	0.174 ± 0.12	-	-	-	0.26 n.s.		
	incongruent	0.199 ± 0.12	-	-	-	-		

The significance level are shown with asterisks (* $p < .01$, *** $p < .001$, n.s. no significant).

Table 3. The differences of the oxy-Hb concentration between incongruent and congruent

task	kana				kanji				2 × 4 ANOVA F-value			
	area	1	2	3	4	1	2	3	4	task (kana · kanji)	area (1 · 2 · 3 · 4)	task × area
The difference of the [oxyHb] (mmol · mm)	0.085 ± 0.13	0.016 ± 0.10	0.107 ± 0.023	0.023 ± 0.013	0.048 ± 0.017	0.010 ± 0.012	0.038 ± 0.018	0.008 ± 0.011				
area		t-value										
1	-	0.069***	0.023	0.062**	-	0.038	0.010	0.040		4.410*	14.282***	3.153*
2	-	-	0.092***	0.007	-	-	0.028	0.002				
3	-	-	-	0.085**	-	-	-	0.030				
4	-	-	-	-	-	-	-	-				

The Stroop effect was defined by the subtraction of oxy-Hb (incongruent condition minus congruent condition). F value of 2 way repeated ANOVA showed significantly main effects and interactions. The significance level are shown with asterisks (* $p < .05$, ** $p < .01$, *** $p < .001$).

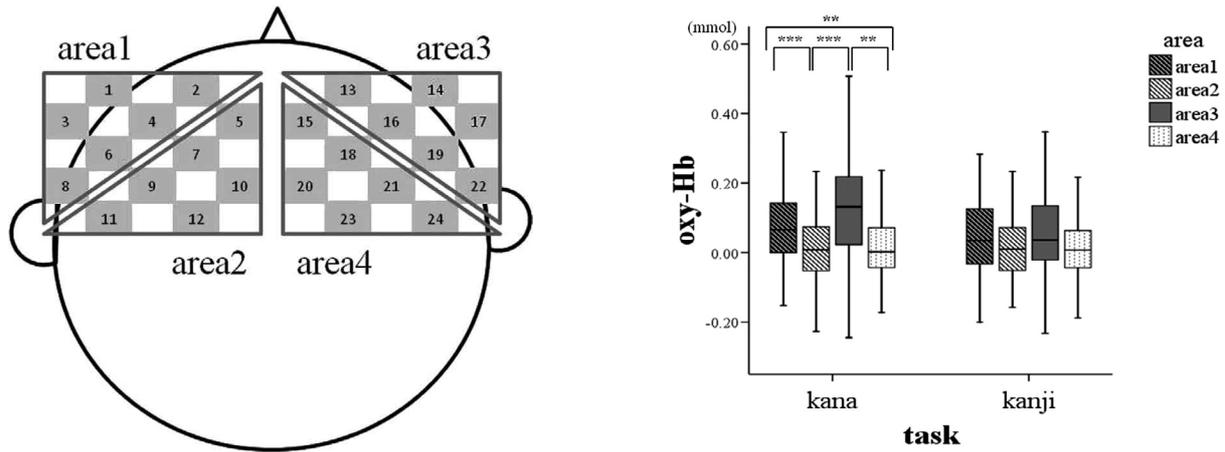


Figure 1. (a) Optodes over the bilateral frontal region. They measured the relative changes in oxyHb and deoxyHb concentrations at 12 measurement points within a 6×6 -cm area of the left and right hemispheres respectively.

(b) The difference of the oxy-Hb concentration under the two conditions (incongruent minus congruent) of four areas in both Kana and Kanji were presented. The difference of the oxy-Hb levels for Area 1 and Area 3 were larger than for Area 2 and Area 4 with Kana whereas there were no significant differences with Kanji. The significance level are shown with asterisks (** $p < .01$, *** $p < .001$).

Table 4. Correlation between oxy-Hb changes elicited by Stroop task and characteristics or mental state of the participants

task	Kana								Kanji							
	congruent				incongruent				congruent				incongruent			
	1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4
BDI	-0.088	0.119	-0.082	0.040	0.039	0.171	0.046	-0.039	-0.004	0.139	0.015	-0.042	0.014	0.129	-0.045	-0.002
STAI-state-	0.136	0.219	0.140	0.091	0.184	0.258	0.169	0.131	0.198	0.217	0.204	0.103	0.074	0.159	0.057	0.025
STAI-trait-	-0.053	0.168	0.075	-0.016	0.016	0.193	0.093	-0.003	0.079	0.187	0.141	0.136	0.038	0.123	0.095	0.045
MOCI	-0.094	-0.150	-0.002	0.051	0.058	0.239	0.165	-0.015	-0.010	0.039	0.087	0.007	0.015	0.008	-0.052	-0.023

DISCUSSION

In this study, we used multi-channel NIRS to investigate the hemodynamic changes elicited with the two Japanese Stroop tasks. We examined the differences in the Stroop effect and hemoglobin changes between Kana and Kanji Stroop tasks. Moreover, we examined whether the participants' psychological conditions or characteristics influenced the hemodynamic changes elicited with the two Japanese Stroop tasks.

In the congruent condition, the participants produced more correct responses in the Kana task than the Kanji task. Although the Stroop effect was observed in both Kana and Kanji tasks, the magnitude of the effect was greater in Kana than in Kanji. Kana is a phonogram script whereas Kanji is an ideogram script. With ideogram scripts, semantic access occurs before phonological processing. Therefore, the Kana incongruent task may cause greater confusion when phonological access to the word occurs automatically and the correct phonological response to the color creates a stronger Stroop effect.

Using NIRS, we examined activation patterns in the prefrontal cortex in individual performing Kana and Kanji versions of the Stroop task. Ehlis *et al.* used the English Stroop task in an NIRS study of the left hemi-frontal area and demonstrated activation of the inferior-frontal regions during the Stroop interference condition (6). The present results showing frontal activation bilaterally in the incongruent conditions both confirm and extend their results. We found significantly larger oxy-Hb increases in the incongruent condition for the Kana task than for the Kanji task. The larger hemodynamic change in the Kana incongruent condition may be related to a larger Stroop effect during the task. It is speculated

that the larger hemodynamic change reflects a stronger conflict with suppressing phonetic reading of Kana.

Area 1 and Area 3, which cover the DLPFC, showed significantly greater activation than Area 2 and Area 4. This result is consistent with previous NIRS studies using the English Stroop task (6). An electroencephalogram (EEG) study found that the ACC and DLPFC are activated and tightly coupled during the Stroop task (17). Therefore, the ACC and the DLPFC may interact with each other to resolve conflict through attentional modulation, and the DLPFC play a critical role in regulating conflict. Using fMRI, Coderre *et al.* reported the Stroop task activated the left inferior parietal lobule during the Kana Stroop task and the left inferior frontal gyrus during the Kanji task. They also reported that the Kana Stroop task activated the frontal areas, precentral gyrus, middle temporal gyrus, and two small regions in the right anterior cingulate gyrus when the Kanji Stroop effect was subtracted from the Kana Stroop effect (incongruent-congruent) (18).

NIRS has a poorer spatial resolution than fMRI and cannot access the deep brain areas. However, NIRS has an advantage in that the original Stroop test can be administered to the participants during measurement. In fMRI or PET studies, participants are restrained in a small space and must respond with a button press rather than speaking. Furthermore, the original Stroop test requires participants to inhibit habitual responses and use selective attention by presenting the 100 color words at the same time. Unlike fMRI or PET studies where the colored words were displayed on the computer screen one by one, NIRS allowed us to use the original style of the Stroop test. We found that a participant's psychological conditions or characteristics, such as depression, anxiety, or obsession, did not influence the hemodynamic

changes observed during the Japanese Stroop tasks. Using NIRS, Taniguchi *et al.* reported that schizophrenia patients showed reduced activation in the prefrontal cortex compared to healthy controls during performance of the Kana Stroop task (19). We suspect that the difference between patients and controls reflects cognitive functioning itself rather than the difference in conditions such as depression, anxiety, or obsession. To accurately assess cognitive functions, it is necessary to remove the influences of the participant's psychological state or characteristics. Therefore, the Japanese Stroop task appears useful for NIRS studies in psychiatry.

To our knowledge, this is the first study to examine the Japanese Stroop task with NIRS. Compared with the Kanji task, the Kana Stroop task showed a greater Stroop effect and hemoglobin changes in the DLPFC. Thus, the Kana Stroop task may be more sensitive and suitable than the Kanji Stroop task. Moreover, we found the psychological conditions or characteristics of a participant did not influence the hemodynamic changes elicited by either Japanese Stroop task. These data suggest that the Kana Stroop task is a useful tool for NIRS study in psychiatric research.

FUNDING

This work was supported by a Grant-in-Aid for Scientific Research from the Japanese Ministry of Education, Culture, Sports, Science, and Technology.

ACKNOWLEDGMENT

We thank all the volunteers who participated in the study.

CONFLICT OF INTEREST

None declared

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