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ORIGINAL ARTICLE

# SIMPLE SMELL IDENTIFICATION TEST USING THREE ODORANTS TO DETECT COGNITIVE DECLINE: INVESTIGATION IN COMMUNITY-DWELLING VOLUNTEERS

Yuki Mikuniya[1)], Shinichi Goto[1)], Akira Sasaki[1)], Reiko Kudo[1)],
Kaori Sawada[2)], Shigeyuki Nakaji[2)], and Atsushi Matsubara[1)]

**Abstract** Background: The number of aging patients with dementia has been increasing in several countries, and a screening method for the early detection of dementia is needed. Objectives: The aim of this study was to evaluate a simple smell identification test for its ability to detect cognitive decline. Subjects and Methods: A total of 659 participants from the 2016 Iwaki Health Promotion Project aged ≧ 40 years were included. Participants underwent a smell identification test using three odorants, namely, India ink, Japanese cypress wood, and curry, out of the Odor Stick Identification Test for the Japanese (OSIT-J) and mini-mental state examination (MMSE) to examine their cognitive function. Results: In analysis of covariance, a statistically significant trend between the age and scores of the three odorants and between the scores of the three odorants and MMSE scores was observed. In multiple regression analysis, the MMSE scores significantly correlated with the scores of the simple smell identification test. From the viewpoint of sensitivity and positive likelihood, the values were not sufficient for screening. Conclusions: A simple smell identification test using three odorants might be useful for the early detection of dementia.

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**Key words:** smell identification test; cognitive decline; mini-mental state examination.

## Introduction

The olfactory nerve, which is the first cranial nerve, originates on the olfactory cells in the nasal mucosa. Its axons enter into the forebrain through the cribriform plate and synapse with the olfactory bulb neurons. The fibers of the lateral olfactory tract rising from the bulbs project to the piriform and entorhinal cortices via the hippocampus, forming a part of the limbic system[1)]. Therefore, anatomical and functional proximity of the brain areas responsible for memory and olfaction can result in a proportional degree of olfaction and memory impairment in patients with cognitive disorders[2)].

Odor identification deficits have been used as first signs of neurodegenerative diseases, such as Alzheimer's disease (AD) and Parkinson's disease (PD), and the relationship between cognitive decline and impaired odor identification has been reported both in Japan and in Western countries[3-9)]. The increasing aging populations and consequent increase in the number of cases with dementia have made the need for a screening method for the early detection of dementia imperative.

The Odor Stick Identification Test for the Japanese (OSIT-J; Daiichi Yakuhin Sangyo, Tokyo, Japan) and the Open Essence (OE; FUJIFILM Wako pure chemical, Osaka, Japan) are used as smell identification tests in Japan, and both tests use the same 12 odorants[10, 11)]. During mass screenings for the detection of mild cognitive impairment (MCI) using smell identification tests,

1) Department of Otorhinolaryngology, Hirosaki University Graduate School of Medicine, Hirosaki, Japan
2) Department of Social Medicine, Hirosaki University Graduate School of Medicine, Hirosaki, Japan
Correspondence: Y. Mikuniya
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an effective test using few odorants would be useful to decrease examination duration and increase screening efficiency.

The purpose of this study was to evaluate the diagnostic ability of a simple smell identification test to detect cognitive decline. The relationship between the cognitive function evaluated by a mini-mental state examination (MMSE) and a smell identification test using three odorants in participants of the Iwaki Health Promotion Project in 2016. The Iwaki Health Promotion Project, which was started in 2005 and is held annually, is a community-based program aimed at improving the average life expectancy by conducting general health check-ups. Three odorants, namely, India ink, Japanese cypress wood, and curry, were chosen from 12 odorants of the OSIT-J according to previous reports[12, 13]. Shiga et al. have shown the usefulness of the curry odorant for olfactory dysfunction screening in Japanese patients and proved that the correct identification of a combination of India ink and Japanese cypress wood is statistically associated with the number of correct answers in tests of 12 odorants[12, 13].

## Subjects and Methods

*Participants*

Residents aged ≧ 20 years living in the Iwaki area, a stable community with a population of approximately 11,000, were invited in the Iwaki Health Promotion Project. As a results 1,148 individuals were participated in 2016. For the relationship between cognitive function and impaired olfactory identification, participants aged ≧ 40 were included in the statistical analysis. The Ethics Committee of Hirosaki University School of Medicine approved the project and data collection for the present study, and all participants signed written informed consents before participating.

*Lifestyles and physical measurements*

Interviews were conducted with participants to gather information regarding age, sex, years of education, present and past medical histories, and smoking and drinking habits. Further, body mass index (BMI; $kg/m^2$) was calculated by measuring participants' heights and weights.

*Assessment of olfactory identification*

The ability to identify odors using three odorants of the OSIT-J, namely, India ink, Japanese cypress wood (*hinoki*), and curry, was assessed. Briefly, the examiner painted each odorant on a thin paraffin paper and passed it to participants. Participants sniffed the paper and were asked to identify the smell by selecting one of six available choices, including four odor names (one of which was correct) and two other possibilities ("unknown smell" and "no smell detected"). The number of correct choices was taken as the score of the smell identification test (0–3 points).

*Assessment of cognitive impairment*

MCI was detected in individuals with scores ≦ 27 points on the MMSE[14]. The examination assesses orientation of time and place; presence or absence of aphasia, apraxia, and agnosis; short and long memory; and calculating ability. The full score is 30 points, and scores decline in proportion with participants' cognitive functions. A clinical psychologist conducted one-to-one examinations in an isolated space and without any time limit.

*Statistical analysis*

Analysis of covariance (ANCOVA) and single regression analysis were conducted to analyze associations between age and the score of the smell identification test by gender. We adjusted covariates: current and past smoking and drinking history and BMI for ANCOVA. In order to examine the effect of smell identification test scores on MMSE scores by gender ANCOVA, multiple regression analysis, and single

**Table 1** Characteristics of participants.

| | Men (n=241) | Women (n=418) | *p*-value |
|---|---|---|---|
| Age (years) | 57.9 ±11.2 | 59.9 ±11.2 | 0.025[a] |
| 40-49 | 66 (27.4%) | 96 (39.8%) | |
| 50-59 | 68 (28.2%) | 100 (41.5%) | |
| 60-69 | 74 (30.7%) | 138 (57.3%) | |
| 70-79 | 26 (10.8%) | 70 (29.0%) | |
| 80+ | 7 (2.9%) | 14 (5.8%) | |
| Education (years) | 12.0 ±1.9 | 11.8 ±1.8 | 0.412[a] |
| Smoking (%) | 59.3 | 22.2 | < 0.001[b] |
| Drinking (%) | 79.7 | 35.2 | < 0.001[b] |
| BMI (kg/m2) | 23.8 ±2.9 | 22.7 ±3.2 | < 0.001[a] |

Values are expressed as mean ± standard deviation.
BMI: body mass index, a: Mann-Whitney U test, b: chi-square test.

regression analysis were used. We adjusted covariates: age, years of education, current and past smoking and drinking history and BMI for ANCOVA and multiple regression analysis. The SPSS 22.0J software (IBM, Armonk, NY, USA) was used for data analysis. $p < 0.05$ was considered statistically significant.

*Sensitivity, specificity, and positive likelihood ratio*

Sensitivity and specificity for the detection of MCI (≦ 27 points on MMSE) with threshold score 0 (0 point), 1 (0 and/or 1 point), and 2 (0-2 points) of simple smell identification test using three odorants was calculated. Further, the positive likelihood ratio (LR) was calculated to determine the cut-off value score for the detection of MCI.

## Results

*Characteristics of participants*

In this study, 898 participants who were ≧ 40 years were enrolled, and 20 participants were excluded from statistical analyses because of missing data and 219 participants because of a history of malignant tumor, cardiovascular disease, stroke, diabetes, or rhino-sinus disease. Data for the remaining 659 participants (241 men and 418 women) were analyzed. Mean ages were 57.9 ± 11.2 years for men and 59.9 ± 11.2 years for women (Table 1). No statistically significant difference was found for years of education. Age and BMI of men were significantly higher than those of women ($p$ = 0.025 and p < 0.001, respectively: U-test), and smoking and drinking habits were significantly higher in men than in women ($p$ < 0.001: chi-square test). Further, 24 (10.0%) men and 26 (6.2%) women had MMSE scores of ≦ 27.

*Association between smell identification test scores and age, or the MMSE*

As a result of ANCOVA, there was significant negative correlation between scores for the three odorants and age in men (Figure 1A) and women (Figure 1B). Mean smell identification scores decline with age significantly (trend $p$ values of < 0.001 in both genders). Linear correlation was obtained by single regression analysis (β-coefficients of -0.018 and -0.032, in men and women, respectively). Values are adjusted for smoking habits, alcohol habits, and BMI.

The MMSE scores in both genders (men: Figure 2A, and women: Figure 2B) were significantly correlated with smell identification test scores (trend $p$ values of 0.025 and 0.004, in men and women, respectively). Linear correlation was obtained by single regression analysis (β-coefficients of 0.485 and 0.348, in men and women, respectively). Values are adjusted for

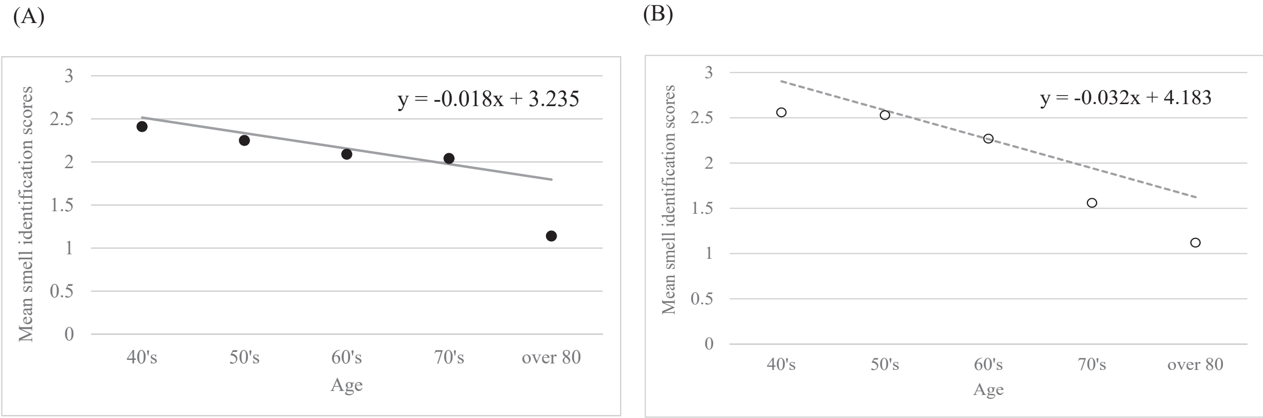


**Figure 1.** Associations between the scores of test using three odorants and participants' age in men (A) and women (B).

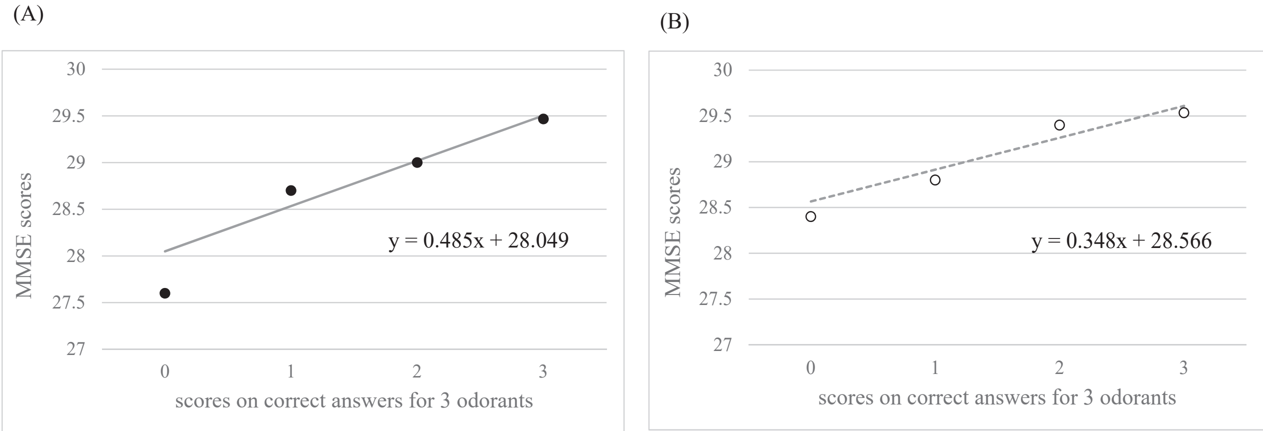


**Figure 2.** The mean MMSE scores for each smell identification score in men (A) and women (B).

**Table 2** Multiple regression analysis with MMSE as a dependent variable.

| | Men | | Women | |
|---|---|---|---|---|
| | β-coeffient | *p* value | β-coeffient | *p* value |
| India ink | 0.130 | **0.025** | 0.058 | 0.218 |
| *hinoki* | 0.069 | 0.256 | 0.113 | **0.017** |
| curry | 0.079 | 0.188 | 0.067 | 0.162 |
| India ink + *hinoki* | 0.128 | **0.030** | 0.118 | **0.015** |
| India ink + curry | 0.139 | **0.018** | 0.079 | 0.104 |
| *hinoki* + curry | 0.098 | 0.110 | 0.125 | **0.011** |
| 3 odorants | 0.140 | **0.019** | 0.125 | **0.012** |

Age, years of education, current and past smoking history, current and past drinking habits, and BMI were adjusted. Significant results ($p < 0.05$) are shown in bold letters.

age, years of education, smoking habits, alcohol habits, and BMI.

In multiple regression analysis (Table 2), when considering a single odorant, the MMSE scores were significantly correlated with the correct answer for the Indian ink only in men and for the Japanese cypress wood only in women. The correct answer for curry did not correlate with the MMSE scores in both genders. When considering combined scores of multiple odorants,

**Table 3** Sensitivity, specificity, and positive LR of test using three odorants.

| Threshold | Sensitivity (%) | Specificity (%) | Positive LR |
|---|---|---|---|
| 0 | 19.2 | 95.1 | 3.9 |
| 1 | 42.3 | 83.4 | 2.5 |
| 2 | 75.0 | 49.1 | 1.5 |

statistically significant associations were found between the MMSE scores in both genders combining scores of Indian ink and Japanese cypress wood and also combining scores of all three odorants.

*Sensitivity, specificity, and positive likelihood ratio of combined scores of identification tests of three odorants to detect MCI*

The specificity and positive LR were the highest in cases with the threshold score 0 among the three categories, but the sensitivity was only 19.2%. The sensitivity was the highest in cases with the threshold score 2, but the resulting LR was only 1.5. The sensitivity and specificity were reasonably high in cases with threshold score 1, but the LR was 2.5 (Table 3).

## Discussion

Cases of age-related dementia, such as AD, are increasing with aging societies. Effective treatment for dementia has not been established, but the detection of MCI during early stages is important to prevent its transition to AD. Olfactory function is impaired with aging and is closely related to cognitive dysfunction[2, 4, 6, 9]. Moreover, Wilson et al. reported that impaired smell identification can predict not only the transition from MCI to AD but also the transition from normal cognition to MCI[4]. Also, a meta-analysis supported the reliability of smell tests as biomarkers for the diagnosis of AD in the preclinical stage[15].

In this study, the diagnostic ability of a simple smell identification test using three odorants was investigated to detect cognitive decline in local residents who participated in the Iwaki Health Promotion Project in 2016. Participants included those aged ≧ 40 years because amyloid-beta deposition pathology in AD is believed to be present from 20 to 30 years before the onset of symptoms[16].

According to our results, the mean score of the test using three odorants significantly decreased with aging, and the decrease in the smell scores was significantly correlated with the decrease in the MMSE scores. Moreover, significant associations were found between the MMSE scores and correct identification of two (Indian ink + Japanese cypress wood) or three (Indian ink + Japanese cypress wood + curry) odorants in both genders. Our findings indicate that a simple smell identification test using few odorants may be useful for the diagnosis of MCI. Shiga et al. have already proven the usefulness of curry odorant testing to screen Japanese individuals for olfactory dysfunction[12]. However, the curry odorant alone is inadequate to detect cognitive function because our multiple regression analysis with MMSE failed to identify a significant association between the MMSE and curry odorant scores. All three odorants chosen for our smell identification test are familiar to Japanese individuals. Indian ink and Japanese cypress wood are familiar smells during school and daily life. In contrast, curry is the most popular food, in Japan. Because the curry odor is considered an appetite stimulator (a primal reaction), it may not be useful for the detection of MCI by itself.

A threshold score of 0 when using the three odorants (Indian ink + Japanese cypress wood

+ curry) displays the highest specificity and LR for the detection of MCI, but the sensitivity is the lowest of all possible odorant combinations. Although the sensitivity of a score 1 when using tests with three odorants was higher that of a score of 0 and the specificity was sufficiently high, the positive LR was not sufficient. Therefore, tests with three odorants seem to be refined before being used as screening tests to detect cognitive decline.

Double et al. showed that five odors (gasoline, banana, pineapple, smoke, and cinnamon) of the University of Pennsylvania 12-item Brief Smell Identification Test are required to adequately discriminate patients with PD from controls[17]. Recently, Umeda-Kameyama et al. showed that six odorants (perfume, rose, Japanese cypress wood, curry, India ink, and gas leak odors) of the OSIT-J were significantly better indicators of cognitive status than the six remaining items by comparing individuals with AD and MCI and normal subjects[18]. The goal of our study was to establish a simple smell identification test using fewer odorants as a screening method to find subjects with subclinical cognitive dysfunction among local residents. To that end, it will be necessary to consider adding one or two odorants, such as a perfume and/or rose, to our simple smell identification test.

In conclusion, the combination of India ink, Japanese cypress wood, and curry significantly correlated with the MMSE scores among both men and women. However, the sensitivity and positive LR were not high enough to ensure its reliability, regardless of the fact that the specificity was reasonable. Further studies are needed to find reliable combinations and numbers of odorants for a screening test to detect cognitive decline.

## Conflicts of interest

The authors declare having no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

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## References

1) Biella G, de Curtis M. Olfactory inputs activate the medial entorhinal cortex via the hippocampus. J Neurophysiol. 2000;83:1924-31.

2) Ottaviano G, Frasson G, Nardello E, Martini A. Olfaction deterioration in cognitive disorders in the elderly. Aging Clin Exp Res. 2016;28:37-45.

3) Doty RL, Bromley SM, Stern MB. Olfactory testing as an aid in the diagnosis of Parkinson's disease: development of optimal discrimination criteria. Neurodegeneration. 1995;4:93-7.

4) Wilson RS, Schneider JA, Arnold SE, Tang Y, Boyle PA, Bennett DA. Olfactory identification and incidence of mild cognitive impairment in older age. Arch Gen Psychiatry. 2007;64:802-8.

5) Haehner A, Hummel T, Reichmann H. Olfactory loss in Parkinson's disease. Parkinson's Dis. 2011. doi:10.4061/2011/450939.

6) Devanand DP, Lee S, Manly J, Andrews H, Schuph N, Doty RL, Stern Y, et al. Olfactory deficits predict cognitive decline and Alzheimer dementia in an urban community. Neurology. 2015; 84:182-9.

7) Growdon ME, Schultz AP, Dagley AS, Amariglio RE, Hedden T, Rentz DM, Johnson KA, et al. Odor identification and Alzheimer disease biomarkers in clinically normal elderly. Neurology. 2015; 84:2153-60.

8) Iijima M, Kobayakawa T, Saito S, Osawa M,

Tsutsumi Y, Hashimoto S, Iwata M. Smell identification in Japanese Parkinson's disease patients: using the odor stick identification test for Japanese subjects. Inter Med. 2008;47:1887-92.

9) Makizako M, Makizako H, Doi T, Uemura K, Tsutsumimoto K, Miyaguchi H, Shimada H. Olfactory identification and cognitive performance in community-dwelling older adults with mild cognitive impairment. Chem Senses. 2014;39:39-46.

10) Kobayashi M, Nishida K, Nakamura S, Oishi M, Shiozaki T, Majima Y, Maeda T, et al. Suitability of the odor stick identification test for the Japanese in patients suffering from olfactory disturbance. Acta Otolaryngol Suppl. 2004:74-9.

11) Nishida K, Kobayashi M, Ogihara H, Takeo T, Kitano M, Takeuchi K. [Clinical usefulness of smell identification test card: Open Essence]. Nihon Jibiinkoka Gakkai Kaiho. 2010;113:751-7. Japanese.

12) Shiga H, Toda H, Kobayakawa T, Saito S, Hirota K, Tsukatani T, Furukawa M, et al. Usefulness of curry odorant of odor stick identification test for Japanese in olfactory impairment screening. Acta Otolaryngol Suppl. 2009:91-4.

13) Shiga H, Yamamoto J, Kitamura M, Nakagawa H, Matsubara T, Seo A, Miwa T. Combinations of two odorants of smell identification test for screening of olfactory impairment. Auris Nasus Larynx. 2014;41:523-7.

14) Saxton J, Morrow L, Eschman A, Archer G, Luther J, Zuccolotto A. Computer assessment of mild cognitive impairment. Postgrad Med. 2009; 121:177-85

15) Kotecha AM, Correa ADC, Fisher KM, Rushworth JV: Olfactory dysfunction as a global biomarker for sniffing out Alzheimer's disease: A meta-analysis. Biosensor. 2018;8:41.

16) Jack CR Jr, Knopman DS, Jagust WJ, Petersen RC, Weiner MW, Aisen PS, Shaw LM, et al. Update on hypothetical model of Alzheimer's disease biomarkers. Lancet Neurol. 2013;12:207-16.

17) Double KL, Rowe DB, Hayes M, Chan DK, Blackie J, Corbett A, Joffe R, et al. Identifying the pattern of olfactory deficits in Parkinson disease using the brief smell identification test. Arch Neurol. 2003;60:545-9

18) Umeda-Kameyama Y, Ishii S, Kameyama M, Kondo K, Ochi A, Yamasoba T, Ogawa S, et al. Heterogeneity of odorant identification impairment in patients with Alzheimer's Disease. Sci Rep. 2017; 7:4798.