



# Does dual-tasking provide additional value in timed “up and go” test for predicting the occurrence of falls? A longitudinal observation study by age group (young-older or old-older adults)

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## Abstract

**Background** Previous studies using relatively large samples and longitudinal observational designs reported dual-tasking had additional value in timed “up and go” test (TUG) for falls assessment among well-functioning older adults.

**Aim** To elucidate the additional value of dual-tasking in TUG for predicting the occurrence of falls among community-dwelling older adults by age group using a predictive model.

**Methods** This longitudinal observation study included 987 community-dwelling older adults at baseline. A TUG without performing another task (single-TUG) and a TUG while counting aloud backward from 100 were conducted at baseline. We computed the dual-task cost (DTC) value, which is used to quantify trends in subjects’ execution of motor tests under dual-task conditions. Data on fall history were obtained using a self-administered questionnaire at the 1-year follow-up. The final analysis included 649 individuals divided into a young-older adult group (aged 60–74 years) and an old-older adult group (aged  $\geq 75$  years). Associations between the occurrence of falls and TUG-related values were analyzed by age group using multivariate logistic regression models.

**Results** For old-older adults, there were significant associations between the occurrence of falls and single-TUG time (odds ratio [OR] 1.143, 95% confidence interval [CI] 1.018–1.285) and DTC value (OR 0.981, 95% CI 0.963–0.999). No significant associations were observed for young-older adults.

**Conclusions** Slower single-TUG time and lower DTC value are associated with the occurrence of falls among old-older adults but not among young-older adults. Dual tasking may provide an additional value in TUG for predicting falls among old-older adults.

**Keywords** Community-dwelling older adults · Dual tasking · Fall · Timed “up and go” test

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## Introduction

The dual-task paradigm is an experimental method used to estimate how the brain deals with two competing tasks performed simultaneously [1]. In general, individuals need to deal with multiple tasks at the same time in daily life, and falls may occur in such situations [2]. Many studies have investigated the usefulness of a dual-task motor performance test to predict the occurrence of falls among community-dwelling older adults. However, an important gap is a review study that summarizes the additional predictive value of dual-tasking in predicting falls in this population [3–9]. As the change related to dual-tasking depends on the type of primal motor task, we should carefully judge the value of dual-tasking for all types of primal motor tests [10].

The timed “up and go” test (TUG), which incorporates basic mobility components (i.e., standing up, walking, turning, and sitting) is widely used for measuring risk for falls among older adults [11, 12]. Some studies have investigated the association between the occurrence of falls and TUG time in the dual-task condition [5, 6, 13–17]. However, previous studies showed inconsistent results and had some methodological issues such as cross-sectional designs or small sample sizes; therefore, a well-designed study using a longitudinal observational design and a large sample is needed to elucidate the additional value of dual-tasking in TUG for predicting the occurrence of falls [5, 6, 13–17]. The components of TUG (standing, walking, turn, walking, and sitting) mean the test is a complicated and difficult motor task for older adults compared with walking. Therefore, this test has potential to reveal the additional value of dual-tasking for predicting the occurrence of falls.

The dual-task effect on a primal task tends to become apparent as a person ages, especially among older adults aged 75 years or older (old-older adults) [18]. This may be because old-older adults have significantly lower physical functions compared with older adults who are younger than 75 years (young-older adults) [19, 20]. This suggests that old-older adults need to allocate and shift their attention to motor tasks more than young-older adults. Consequently, dual-tasking may affect old-older adults more. The usefulness of a dual-task motor performance test for predicting falls may depend on the person’s age group (young-older and old-older adults), and any association may be clearer among old-older adults than young-older adults. However, no studies have investigated this hypothesis.

Given the age-related decline in cognitive and physical performance, we hypothesized that a dual-task TUG may be useful to predict the occurrence of falls, especially

among individuals classified as old-older adults. The present longitudinal observation study aimed to elucidate the additional value of dual-tasking in TUG for predicting the occurrence of falls among Japanese community-dwelling older adults by age group: young-older or old-older adults. Dual-task cost (DTC) is used to quantify trends in subjects’ execution of motor tests under dual-task conditions [4, 5, 21]. It is computed by comparing performance on single- and dual-tasks to determine the change in performance under the dual-task condition [4, 5, 21]. In the present study, we used the DTC as a score of dual-task performance.

## Methods

### Participants

The present study used a longitudinal observation design. We visited 20 community centers in Inami town, Hyogo prefecture, Japan, from April to October 2016 and recruited 1039 adults who lived independently in the community. The inclusion criteria were: (1) aged 60 years older and (2) able to walk independently with or without an assistive device. The exclusion criteria were: (1) unable to perform the single- or dual-TUG; (2) incomplete data on any of the measurements; and (3) self-reported neurological disorders that could affect mobility or balance. The final baseline sample comprised 987 participants. At the 1-year follow-up, 322 participants had dropped out and six participants had missing data for falls and Rapid Dementia Screening Test (RDST). The final analytical sample comprised of 649 individuals (follow-up rate: 649/987, 66%). In total, 331 individuals were classified as young-older adults (aged 60–74 years), and 318 were classified as old-older adults (aged  $\geq 75$  years).

### Sociodemographic measures and occurrence of falls in the 1-year follow-up

Data on sociodemographic characteristics were collected using a self-administered questionnaire at baseline. Questionnaire items included age, sex, height, weight, and medical history (hypertension, cardiovascular disease, respiratory disease, diabetes mellitus, and osteoarthritis). A fall was defined as “an event that resulted in the participant unintentionally coming to the ground or other lower level.” [22] Data regarding the occurrence of falls during the 1-year follow-up were collected using a self-administered questionnaire at the follow-up measurement and were used as a main outcome in the present study. Cognitive function was assessed using the RDST at baseline and after the 1-year follow-up [23, 24].

## TUG

Participants were asked to stand up from a seated position in an armchair, walk 3 m, turn around, walk back to the chair, and sit down again (single-TUG) [12]. Next, they were asked to complete the TUG while counting backward (by one) aloud from 100 (dual-TUG). We chose counting backwards (by one) as an additional cognitive task for the two following reasons: (1) its dual-tasking effects have been observed in previous studies; and (2) most older adults with normal cognitive function can perform this task without extensive instruction, which was clinically important for smooth completion of the test given the number of participants included in this study [25, 26]. No instructions were given regarding which task to prioritize during the dual-TUG. Before measurements were taken, we explained how to perform the dual-TUG to ensure participants understood the procedures correctly. This arithmetic count task is easy to perform, and some individuals are able to count rhythmically through familiarity with the task. Repetition of the task could result in familiarity that weakens its dual-tasking effect. Therefore, we instructed participants not to perform this cognitive task too many times before the dual-TUG was conducted. The times taken to complete the single- and dual-TUG were measured using a digital stopwatch. Participants wore their usual footwear and were asked to walk at a comfortable and safe pace. One trial was performed in each condition.

The last number spoken during the dual-TUG by each participant was also recorded. We calculated participants' backward counting speed during the dual-TUG. Backward counting speed (n/s) was equal to the difference between 100 minus the last number counted during the dual-TUG divided by the time taken to complete the dual-TUG. To assess trends in participants' execution of the motor test under the dual-task condition, we used the below formula to compute the DTC [4, 5, 21]. We also used an arithmetic task as a secondary task. A previous review that summarized the usefulness of DTC using an arithmetic task as a secondary task showed the task had no usefulness for predicting falls in older adults [8]. However, as the primal task in that study was walking and not the TUG, the value of an arithmetic task as a secondary task remains unclear.

$$\text{DTC (\%)} = 100 \times (\text{dual-TUG time} - \text{single-TUG time}) / ((\text{single-TUG time} + \text{dual-TUG time}) / 2).$$

Higher DTC values indicated longer completion times under the dual-task condition than under the single-task condition, and lower DTC values indicated smaller differences in performance between the single- and dual-task conditions [4, 5, 21].

## Statistical analysis

The normal distribution of continuous variables was confirmed by the results of a Shapiro–Wilk test. We also confirmed the normality of the distribution by observing the histogram and QQ-plot. Demographic characteristics in each age group were compared between participants who had experienced falls during the follow-up year and those who had not experienced falls using unpaired *t* tests or likelihood tests. Single- and dual-TUG time and backward counting speed in the dual-TUG were compared between participants who had experienced falls during the follow-up year and those who had not experienced falls using a Mann–Whitney *U* test. The DTC value was compared between these groups using an unpaired *t* test. Single regression analysis was conducted to investigate the association between the single-TUG time and age; this was to confirm the rapid-change in TUG time with ageing. Multivariate logistic regression analysis was then used to investigate associations between the occurrence of falls in the follow-up year and single-TUG time and DTC value by age group (Model 1). Age, sex, height, and weight were included as covariates in each regression model. In the second logistic regression model (Model 2), we added RDST score at baseline, the change in RDST score (difference in RDST score between baseline and after the 1-year follow-up) to reflect cognitive decline and backward counting speed as covariates. In the final logistic regression model (Model 3), we also added comorbidities (hypertension, cardiovascular disease, respiratory disease, diabetes, and osteoarthritis) as covariates.

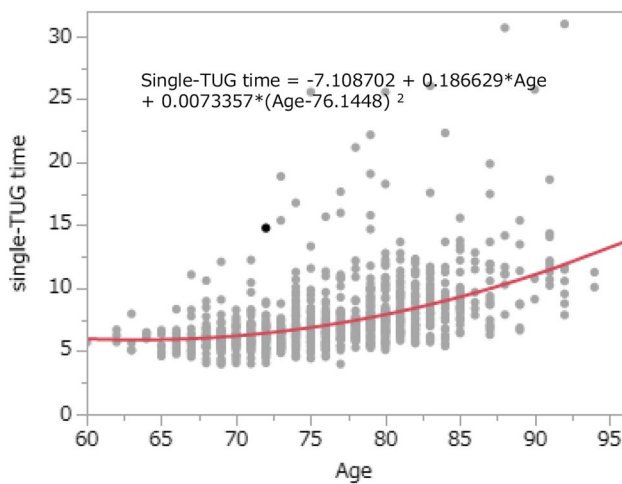
## Results

Participants' characteristics are summarized in Table 1. Among young-older adults, 253 individuals (76%) were classified as non-fallers and 78 (24%) as fallers. Among the old-older adults, 221 individuals (69%) were non-fallers and 97 (31%) were fallers. There were no significant differences in participants' characteristics at baseline between the non-fallers and fallers in either age group. The regression curve for single-TUG time and age is shown in Fig. 1 (single-TUG time =  $-7.108702 + 0.186629 \times \text{age} + 0.0073357 \times (\text{age} - 76.1448)^2$ ). Table 2 shows the comparisons of TUG-related values (DTC value, single-TUG time, dual-TUG time, and backward counting speed in the dual-TUG) between non-fallers and fallers in both age groups. In young-older adults, fallers took longer to complete the single-TUG than non-fallers ( $p = 0.025$ ). In old-older adult group, fallers showed smaller DTC values than non-fallers ( $p = 0.005$ ). In other words, fallers showed a smaller difference between the single- and dual-task conditions than non-fallers. The results of the multivariate logistic regression analyses for young-older

**Table 1** Participants' characteristics at baseline

	Young-older adults			Old-older adults		
	Non-fallers ( <i>n</i> =253)	Fallers ( <i>n</i> =78)	<i>p</i> value	Non-fallers ( <i>n</i> =221)	Fallers ( <i>n</i> =97)	<i>p</i> value
Age (years)	71.7±2.8	72.1±2.9	0.289	80.8±3.9	80.4±3.6	0.289
Sex (female) <i>n</i> (%)	164 (65)	53 (68)	0.610	139 (63)	69 (71)	0.152
Height (cm)	155.6±7.5	155.4±9.9	0.861	153.2±9.2	152.6±8.2	0.624
Weight (kg)	55.9±10.0	57.4±12.1	0.271	54.0±9.7	54.5±8.2	0.704
Hypertension <i>n</i> (%)	109 (43)	39 (50)	0.284	117 (53)	58 (60)	0.257
Cardiovascular disease <i>n</i> (%)	19 (8)	8 (9)	0.679	29 (13)	16 (16)	0.432
Respiratory disease <i>n</i> (%)	11 (4)	5 (6)	0.472	7 (3)	3 (3)	0.972
Diabetes <i>n</i> (%)	32 (13)	11 (14)	0.740	26 (12)	16 (16)	0.259
Osteoarthritis <i>n</i> (%)	9 (4)	7 (9)	0.068	24 (11)	9 (9)	0.667
Rapid Dementia Screening Test	11.0±1.7	10.6±2.5	0.116	9.1±2.9	9.3±2.9	0.561

Values are means ± standard deviations or *n* (%)

**Fig. 1** Association between timed “up and go” test time and age in community-dwelling older adults

adults are shown in Table 3. In Model 1, single-TUG time was significantly associated with the occurrence of falls in the follow-up year (odds ratio [OR] 1.143, 95% confidence interval [CI]: 1.012–1.292,  $p=0.032$ ). However, after adding covariates (RDST and backward counting speed during the dual-TUG) into Model 1, the association was no longer significant. The results of the multivariate logistic regression analyses for old-older adults are shown in Table 4. In Model 1, single-TUG time (OR 1.111, 95% CI 1.008–1.224,  $p=0.034$ ) and DTC value (OR 0.979, 95% CI 0.962–0.996,  $p=0.017$ ) and were significantly associated with the occurrence of falls in the follow-up year. These significant associations remained after adding covariates (RDST at baseline, change in RDST score, and backward counting speed during the dual-TUG) into Model 1 (single-TUG time, OR 1.124, 95% CI 1.002–1.261,  $p=0.045$ ; DTC value, OR 0.980, 95% CI 0.962–0.998,  $p=0.029$ ). Furthermore, these significant

associations remained even after adding comorbidities (hypertension, cardiovascular disease, respiratory disease, diabetes, and osteoarthritis) as covariates (single-TUG time: OR 1.143, 95% CI 1.018–1.285,  $p=0.024$ ; DTC value: OR 0.981, 95% CI 0.963–0.999,  $p=0.049$ ). The OR indicates the change in odds of falling by a one-unit increase in the exploratory variable.

## Discussion

In the present study, single- and dual-TUG were administered to community-dwelling older adults at baseline. Information regarding the occurrence of falls was obtained via a self-administered questionnaire at the 1-year follow-up measurement. We first ascertained the trend of declining TUG time with increasing age. Next, we investigated associations between the occurrence of falls and single-TUG time and DTC values for each age group (young-older or old-older adults) to elucidate the additional value of dual-tasking in the TUG for predicting the occurrence of falls.

Our results showed that the time needed to perform the TUG increased rapidly after age 75 years. This result was consistent with previous studies [19, 20, 27]. In the early phase of ageing, comprehensive physical function, which can be assessed by a general motor test such as the TUG, is maintained a certain level in Japanese individuals [27]. However, even among well-functioning individuals, rapid changes in physical function may occur after the age of 75 years. This suggests that older adults aged  $\geq 75$  years have a potential risk for decline in physical function.

The results of the logistic regression analysis (Table 3) showed no significant association between the occurrence of falls and DTC value in young-older adults. This result supports our hypothesis and indicates that dual-tasking may not provide additional value in TUG for predicting the

**Table 2** Single- and dual-timed “up and go” test time, dual-task cost value, and backward counting speed in the dual-timed “up and go” test among non-fallers and fallers

	Young-older adults			Old-older adults		
	Non-fallers (n=253)	Fallers (n=78)	p value	Non-fallers (n=221)	Fallers (n=97)	p value
DTC value (%)	15.4 ± 15.5	14.0 ± 15.0	0.491	16.8 ± 16.5	11.3 ± 13.6	0.005
Single-TUG time (s)	6.4 ± 1.6	7.1 ± 2.9	0.020	7.9 ± 2.4	8.7 ± 3.8	0.140
Dual-TUG time (s)	7.5 ± 2.1	8.2 ± 4.0	0.145	9.5 ± 3.0	9.8 ± 4.3	0.893
Backward counting speed in the dual-TUG (n/s)	1.3 ± 0.3	1.2 ± 0.3	0.190	1.0 ± 0.4	1.0 ± 0.3	0.460

Values are means ± standard deviations. Non-fallers were older adults with no history of falling in the previous year. Fallers were those with a history of falling in the previous year

Backward counting speed during dual-TUG = the number reached while counting backward from 100 during the dual-TUG/the dual-TUG time

Single- and dual-TUG time and backward counting speed in the dual-TUG were compared between non-fallers and fallers using a Mann–Whitney *U* test; DTC values were compared between the groups using an unpaired *t* test

*DTC* dual-task cost,  $DTC = (dual-TUG\ time - single-TUG\ time) / \{(dual-TUG\ time + single-TUG\ time) / 2\}$

*Single-TUG* timed “up and go” test without an additional task, *dual-TUG* timed “up and go” test with an additional task

**Table 3** Associations between timed “up and go” test-related variables and the occurrence of falls in the follow-up year in young-older adults

	Model 1		Model 2		Model 3	
	Odds ratio (95% CI)	p value	Odds ratio (95% CI)	p value	Odds ratio (95% CI)	p value
Single-TUG time	1.143 (1.012–1.292)	0.032	1.094 (0.939–1.275)	0.249	1.088 (0.933–1.270)	0.280
DTC value	0.997 (0.980–1.015)	0.746	0.993 (0.975–1.012)	0.482	0.994 (0.975–1.013)	0.513
Age	1.047 (0.951–1.154)	0.347	1.042 (0.945–1.149)	0.414	1.038 (0.939–1.148)	0.466
Sex (female)	1.649 (0.747–0.639)	0.216	1.815 (0.808–4.078)	0.149	1.716 (0.757–3.889)	0.196
Height	1.000 (0.953–1.049)	0.998	1.001 (0.954–1.051)	0.958	1.001 (0.953–1.051)	0.977
Weight	1.027 (0.995–1.061)	0.102	1.028 (0.995–1.063)	0.095	1.028 (0.992–1.064)	0.129
Backward counting speed during the dual-TUG test	–	–	0.874 (0.314–2.430)	0.797	0.852 (0.302–2.402)	0.762
RDST score at baseline	–	–	0.882 (0.724–1.075)	0.215	0.890 (0.730–1.086)	0.251
Change in RDST score	–	–	0.846 (0.692–1.033)	0.101	0.838 (0.683–1.027)	0.088
Hypertension	–	–	–	–	0.967 (0.544–1.717)	0.909
Cardiovascular disease	–	–	–	–	1.095 (0.410–2.923)	0.856
Respiratory disease	–	–	–	–	1.633 (0.518–5.149)	0.402
Diabetes	–	–	–	–	0.964 (0.441–2.107)	0.926
Osteoarthritis	–	–	–	–	2.215 (0.761–6.445)	0.144

Multivariate logistic regression analyses were conducted with the occurrence of falls during the follow-up year as the dependent variable

Model 1 included TUG-related variables (single-TUG time and DTC) as independent variables, and age, sex, height, and weight as covariates

Model 2 included all variables from Model 1, with RDST score and the change in RDST score added as covariates

Model 3 included all variables from Model 2, with comorbidities (hypertension, cardiovascular disease, respiratory disease, diabetes, and osteoarthritis) added as covariates

*Single-TUG* timed “up and go” test without an additional task, *dual-TUG* timed “up and go” test with an additional task, *DTC* dual-task cost,  $DTC = (dual-TUG\ time - single-TUG\ time) / \{(dual-TUG\ time + single-TUG\ time) / 2\}$ , *RDST* Rapid Dementia Screening Test, *Change in RDST score* the difference of RDST score between baseline and the 1-year follow-up

occurrence of falls among young-older adults. Our results suggested that young-older adults can cope with situations such as a dual-task, or that falls may occur in this age group for other reasons [28]. The results in Table 3 also showed no

significant association between the occurrence of falls and single-TUG time. This suggested that the usefulness of a single-TUG for predicting the occurrence of falls is limited, which is consistent with other studies showing no significant

**Table 4** Associations between timed “up and go” test-related variables and the occurrence of falls in the follow-up year in old-older adults

	Model 1		Model 2		Model 3	
	Odds ratio (95% CI)	<i>p</i> value	Odds ratio (95% CI)	<i>p</i> value	Odds ratio (95% CI)	<i>p</i> value
Single-TUG time	1.111 (1.008–1.224)	0.034	1.124 (1.002–1.261)	0.045	1.143 (1.018–1.285)	0.024
DTC value	0.979 (0.962–0.996)	0.017	0.980 (0.962–0.998)	0.029	0.981 (0.963–0.999)	0.049
Age	0.925 (0.857–0.998)	0.044	0.924 (0.852–1.002)	0.057	0.918 (0.845–0.997)	0.042
Sex (female)	1.494 (0.656–3.403)	0.339	1.460 (0.635–3.356)	0.373	1.595 (0.678–3.747)	0.285
Height	1.006 (0.958–1.056)	0.815	1.004 (0.956–1.055)	0.869	1.009 (0.959–1.061)	0.727
Weight	1.008 (0.973–1.044)	0.671	1.008 (0.972–1.044)	0.673	1.003 (0.966–1.041)	0.894
Backward counting speed during the dual-TUG test	–	–	1.214 (0.449–3.282)	0.702	1.434 (0.522–3.940)	0.485
RDST score at baseline	–	–	1.003 (0.879–1.144)	0.963	0.994 (0.872–1.134)	0.932
Change in RDST score	–	–	0.962 (0.849–1.089)	0.536	0.962 (0.849–1.089)	0.537
Hypertension	–	–	–	–	1.380 (0.820–2.322)	0.225
Cardiovascular disease	–	–	–	–	1.379 (0.682–2.790)	0.371
Respiratory disease	–	–	–	–	0.869 (0.194–3.898)	0.854
Diabetes	–	–	–	–	1.516 (0.727–3.164)	0.267
Osteoarthritis	–	–	–	–	0.736 (0.307–1.762)	0.491

Multivariate logistic regression analyses were conducted with the occurrence of falls during the follow-up year as the dependent variable

Model 1 included TUG-related variables (single-TUG time and DTC) as independent variables, and age, sex, height, and weight as covariates

Model 2 included all variables from Model 1, with RDST score and the change in RDST score added as covariates

Model 3 included all variables from Model 2, with comorbidities (hypertension, cardiovascular disease, respiratory disease, diabetes, and osteoarthritis) added as covariates

*Single-TUG* timed “up and go” test without an additional task, *dual-TUG* timed “up and go” test with an additional task, *DTC* dual-task cost,  $DTC = (\text{dual-TUG time} - \text{single-TUG time}) / \{(\text{dual-TUG time} + \text{single-TUG time}) / 2\}$ , *RDST* Rapid Dementia Screening Test, *Change in RDST score* the difference of RDST score between baseline and the 1-year follow-up

association between the TUG and the occurrence of falls [29]. Interestingly, some studies have shown that even older adults with high physical function frequently fell and were severely injured [30, 31]. These results imply that older adults with high physical function may frequently expose themselves to dangerous conditions and increase their risk for falling [32]. In such cases, the environmental condition becomes an important risk factor, rather than physical function. Our results regarding falls for young-older adults may reflect similar characteristics. However, the present study did not include environmental assessment, and further studies are needed to clarify this issue.

In contrast, the results of the logistic regression analysis for old-older adults (Table 4) showed that single-TUG time and DTC value were significantly associated with the occurrence of falls in this group. This result suggests that the dual-task method may provide additional value in TUG for predicting the occurrence of falls among old-older adults. Importantly, a smaller DTC value (i.e., a smaller difference between dual- and single-TUG time) was significantly associated with the occurrence of falls in this age group. This supported our previous cross-sectional study indicating that lower DTC values were significantly associated with fall history in the transitional-functioning

group, but not in the well-functioning group [5]. There are two possible explanations for the better performance on the dual-task test. First, the fall risk may be lowered because of adaptation to the challenging dual-task situation [10]. Second, the fall risk may increase because individuals cannot ensure their postural stability when slowing gait speed; that is, when using an incorrect postural strategy in the dual-task situation. Individuals who adopt this strategy tend to sacrifice their postural stability in complicated situations [33]. Our results support the latter explanation. Older individuals who are experiencing a decline in physical function may adopt an incorrect postural strategy; consequently, their fall risk may increase. Considering the results for young-older adults and old-older adults together suggests that the usefulness of the dual-task method for predicting the occurrence of falls may differ by age group. Importantly, diverse results have been shown in studies investigating associations between dual-tasking and falls, and the dual-task method should be carefully adapted for the purpose of predicting falls in community-dwelling older adults [3, 4]. In this study, single-TUG time was significantly associated with the occurrence of falls, which was consistent with results reported by previous studies, and indicates that the single-TUG time

is useful for predicting the occurrence of falls among old-older adults [34].

The strengths of our study were that we used a comparatively large sample and a longitudinal design. A novel aspect of this study was that we elucidated the additional value of dual-tasking in TUG for predicting the occurrence of falls in well-functioning older adults by age group. This study had some limitations that should be mentioned. First, the follow-up rate was relatively low (649/987, 66%), and selection bias might have occurred. Second, some data regarding falls during the follow-up period were not obtained, such as the number falls and the circumstances in which participants fell, and we could not conduct sub-analyses to obtain other clinical information. Third, potential confounders may exist, such as comorbidities other than those shown in Table 1 (e.g., diseases that may affect balance and memory, pain, and sedative drug use) [35, 36]. These limitations might have affected our results. Fourth, we did not measure the effect of other types of additional tasks (e.g., manual tasks) on TUG. The effect of an additional task on the primary task differs according to the type of additional task [18, 26]. Additionally, the cognitive task we used might have been easy to perform, and the dual-tasking effect might have been weakened by participants' familiarity.

In conclusion, a slower single-TUG and lower DTC value are associated with the occurrence of falls among old-older adults, but not among young-older adults. Dual tasking may provide an additional value in TUG for predicting falls among old-older adults.

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## Compliance with ethical standards

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

**Human and animal rights** This study was carried out in accordance with the principles of the Declaration of Helsinki, and was approved by the Research Ethics Committee of Kobe Gakuin University (Approval No. HEB100806-1).

**Informed consent** Informed consent was obtained from all individual participants included in the study.

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