Behavior Research Methods (2018) 50:1602–1613
https://doi.org/10.3758/s13428-018-1047-2

New protocol for dissociating visuospatial working memory ability in reaching space and in navigational space

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Published online: 27 April 2018

Abstract
Several studies have demonstrated that the processing of visuospatial memory for locations in reaching space and in navigational space is supported by independent systems, and that the coding of visuospatial information depends on the modality of the presentation (i.e., sequential or simultaneous). However, these lines of evidence and the most common neuropsychological tests used by clinicians to investigate visuospatial memory have several limitations (e.g., they are unable to analyze all the subcomponents of this function and are not directly comparable). Therefore, we developed a new battery of tests that is able to investigate these subcomponents. We recruited 71 healthy subjects who underwent sequential and simultaneous navigational tests by using an innovative sensorized platform, as well as comparable paper tests to evaluate the same components in reaching space (Exp. 1). Consistent with the literature, the principal-component method of analysis used in this study demonstrated the presence of distinct memory for sequences in different portions of space, but no distinction was found for simultaneous presentation, suggesting that different modalities of eye gaze exploration are used when subjects have to perform different types of tasks. For this purpose, an infrared Tobii Eye-Tracking X50 system was used in both spatial conditions (Exp. 2), showing that a clear effect of the presentation modality was due to the specific strategy used by subjects to explore the stimuli in space. Given these findings, the neuropsychological battery established in the present study allows us to show basic differences in the normal coding of stimuli, which can explain the specific visuospatial deficits found in various neurological conditions.

Keywords Visuospatial working memory · Sequential presentation · Simultaneous presentation · Reaching space · Navigational space · Information processing

In the past 20 years, the number of studies that investigated the specific mechanisms underlying visuospatial cognition has greatly increased.

A great contribution to this field was provided by Baddeley and Hitch (1974), who described the visuospatial component of working memory (WM) as the subsystem responsible for maintaining and processing visuospatial information.

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Because of the growing evidence of dissociation between the visual and spatial aspects of visuospatial working memory (VSWM), several authors proposed a distinction in Baddeley's model between a visual component (*visual cache*), dealing with color and shape, and a spatial component (*inner scribe*) for processing spatial locations (Della Sala, Gray, Baddeley, Allamano, & Wilson, 1999; Klauer & Zhao, 2004; Logie, 1995). Supporting this distinction, several studies both in animals and in humans have confirmed this division at the anatomical level by showing that the visual and spatial memory components involve different regions of the brain, namely, the dorsal ("what") and the ventral ("where") pathways. The ventral pathway projects to the inferior temporal lobe and is considered relevant for object identification, whereas the dorsal pathway projects to the parietal cortex and is considered relevant for the processing of spatial information (Courtney, Ungerleider, Keil, & Haxby, 1996; Kravitz, Saleem, Baker, & Mishkin, 2011; Ungerleider & Haxby, 1994; Wilson, 1993).

However, in the past twenty years, other important distinctions have been proposed concerning the architecture of VSWM.

It has been proposed that memory for locations in reaching space must be differentiated from memory for locations in navigational space (route memory) (Ekstrom et al., 2003; Hartley & Burgess, 2005; Hartley, Maguire, Spiers, & Burgess, 2003; Maguire et al., 1998; Piccardi et al., 2008; White & McDonald, 2002). This differentiation is also supported by lesion studies that indicate double dissociations between deficits in near and navigational space (De Renzi, 1982; Piccardi, Iaria, Bianchini, Zompanti, & Guariglia, 2011).

Furthermore, Piccardi et al. (2011) demonstrated that the route-memory system is dissociated from the system that memorizes sequences of locations in near space (Piccardi et al., 2014; Piccardi et al., 2011). This hypothesis is supported by clinical observations of patients who fail to store and recall sequences on the Corsi block-tapping test without any navigational impairment as well as of patients affected by topographical disorientation who perform flawlessly on the Corsi test (Bianchini, Palermo, et al., 2014). Similarly, Palermo and collaborators (2014) described a patient with developmental topographical disorientation who showed specific deficits in navigational tasks without problems in other visuospatial tasks (i.e., the Corsi test; Palermo et al., 2014).

Moreover, studies using neuroimaging techniques demonstrated the existence of a neural network related to route-based navigation (Nemmi et al., 2013) and demonstrated that specific pathologies can alter distinct navigational processes that are subtended by specific brain activity alterations (Palermo et al., 2014). Taken together, these studies provide further support for the theory that there are two distinct visuospatial memory systems for route memory and near memory.

Furthermore, several authors advanced the hypothesis that VSWM material is coded differently according to presentation modality. Specifically, Pazzaglia and Cornoldi (1999) proposed a distinction within VSWM between a simultaneous modality and a sequential modality of stimulus presentation. The simultaneous modality refers to spatial locations presented simultaneously in a working memory task and is related to static processes (Pickering, Gathercole, Hall, & Lloyd, 2001), whereas the sequential modality refers to spatial locations sequentially presented such that the subjects have to recall each previous position, and it is related to dynamic processes (Pickering et al., 2001). Evidence collected from various groups of children and adults suffering from different neurological diseases supports a separation between simultaneous and sequential visuospatial processing (I. C. Mammarella, Borella, Pastore, & Pazzaglia, 2013; I. C. Mammarella, Coltri, Lucangeli, & Cornoldi, 2009; I. C. Mammarella et al., 2006; N. Mammarella, Cornoldi, & Donadello, 2003; Piccardi et al., 2011; Wansard et al., 2015).

Moreover, Lecerf and De Ribaupierre (2005) have proposed that the use of different modes of encoding likely also depends on the type of task. Specifically, the authors distinguished extrafigural encoding responsible for anchoring objects in relation to an external reference frame from intrafigural encoding based on the relationships between different items in a pattern, the latter being further divided into pattern encoding (leading to a global visual image) and path encoding (leading to spatial–sequential position). Hence, pattern encoding is involved when the spatial items are simultaneously presented, whereas path encoding is involved when the spatial items are sequentially presented (Lecerf & De Ribaupierre, 2005).

In light of these considerations, we established a new protocol to investigate both the simultaneous and sequential components of VSWM in navigational and reaching space. Furthermore, to evaluate the influence of the presentation modality on accuracy of the subjects and to account for the different portions of the space, in Experiment 1, we have analyzed VSWM in navigational space while examining both the simultaneous and sequential components of spatial processing using a tool developed ad hoc. It is a sensorized platform named "SMARTile" and managed with dedicated software.

Moreover, we have developed a paper protocol suited to analyzing VSWM in reaching space resembling the simultaneous and sequential characteristics of the navigational tasks.

Tasks were set out with items at increasing levels of difficulty to obtain the span score of each subject in each task. Indeed, the span score has been demonstrated to be a reliable parameter to evaluate individual differences in working memory tasks (Cornoldi & Vecchi, 2003). Additionally, two versions (Version 1 and Version 2) of each experimental task were developed to allow multiple assessments for each subject. In Experiment 1, we tested whether the two versions are indeed equivalent and investigated the factor structure underlying the tasks. In Experiment 2, a pilot study was performed to analyze the influence of spatial sequential and simultaneous processes on the subjects' exploration strategies in both reaching and navigational space. (For a discussion of this issue, see the theory of Pazzaglia & Cornoldi, 1999.)

The neuropsychological battery proposed in the present study allows a complete and exhaustive assessment of VSWM, thus providing clinicians with a new tool to diagnose the specific VSWM deficits linked to different neurological conditions.

Experiment 1

To analyze VSWM in navigational and reaching spaces, considering both the simultaneous and sequential components of visual processing, we developed a new tool named the "**S**ystem for **M**onitoring and **A**ssisted **R**ehabilitation by advanced **Tile**" (SMARTile) for the navigational space analysis and several paper tasks to investigate the same components in reaching space.

Two versions of each experimental task have been developed to allow multiple assessments for each subject.

Method

Subjects

Seventy-one healthy volunteers [M/F: 26/45; mean age: 26.63 (*SD* = 6.84); mean years of education: 16.50 (*SD* = 1.78)] participated in the experiment.

None of the subjects had a history of neurological or mental illness, and all of them were able to walk without aid. All the subjects underwent an evaluation of their intellectual level by means of Raven's Progressive Matrices '47 (Raven, 1949).

All the subjects were recruited from the Department of Psychology at the Sapienza University of Rome. The experimental procedures were approved by the Institutional Review Board of the Department of Psychology. The study has been carried out in accordance with the Code of Ethics of the World Medical Association (Declaration of Helsinki). Written consent was obtained from each subject.

Procedure

The experimental sessions were performed in a quiet room with artificial lighting.

During the navigational tasks, the experimenter was sitting behind the subject, and during the assessment in the reaching space, the experimenter was in front of the subject.

A training trial was performed before each task.

The execution time for each task did not exceed 15 min (when the maximum sequence length was reached). At the end of each task, the subjects rested for 5 min before performing a new task.

All the tasks were performed in a randomized order among the subjects.

Experimental paradigms

Navigational space In navigational space, spatial–sequential and spatial–simultaneous working memories were assessed using an innovative tool, a sensorized platform named SMARTile, provided with software developed ad hoc that can be installed on any notebook computer and connected via wireless Internet to a sensorized platform.

The platform is made up of 25 semirigid plastic "smart" tiles (each with dimensions 50 cm × 50 cm) with an electronic component through which the software records changes in the body positions of subjects (Fig. 1a).

The platform can provide different types of visual feedback via blue, red, or green light-emitting diodes (LEDs; Fig. 1b) and/or acoustic feedback.

The characteristics of the apparatus have made it possible to meet specific criteria, such as the control of the activities of each tile independently. This characteristic allows for building different combinations of trials. When the software starts, the experimenter can verify the correct functioning of each tile through the graphical interface of the platform (Fig. 1c and d) and can create new or modify existing experimental paradigms.

The particular shape of the tiles allows for the creation of a smooth and continuous surface, similar to a carpet, on which the subject walks (total platform: 250 cm × 250 cm).

The particular configuration of the tiles does not allow for the subject to have any cue within the platform except for the boundaries within the platform, thus eliminating possible distractors and/or facilitators in solving the tasks.

An intrinsic limit of the instrument is the necessity that the subject be able to walk independently without any particular difficulty in maintaining balance. This limit is linked to the peculiarities of the instrument, the use of which requires adherence to three rules: (I) stand with both feet on the indicated tile, (II) do not keep your feet on two tiles, and (III) the next step must be performed approximately 1 s after the previous one.

The particular configuration of the SMARTile system and the specific features of the tiles enabled us to build various experimental paradigms that differed in terms of sequential or simultaneous encoding load.

Specifically, the following tasks were used:

- *Motor control test (MCT)* Subjects were enrolled in the study only if they were able to perform this task. Visual stimuli were presented through blue lights. For each tile, the light stayed on until the subject moved to it. The lights changed one by one until the end of a nine-tile sequence. In this case, the subject was not required to remember the path just taken.
- *Sequential Route A (SRA)* Visual stimuli were presented through blue lights that appeared on the platform according to a precise space–time sequence (Fig. 2a). Each stimulus remained visible until the last light of the sequence had been presented. After a few seconds, all the visual stimuli simultaneously disappeared. Then the subject heard a sound (lasting 2 s) that indicated he or she should stand on the platform and repeat the same sequence just observed, walking on the tiles previously lit. Sequences of increasing length (from three to nine tiles) were presented until the subject failed to reproduce two out of three trials of a given length.

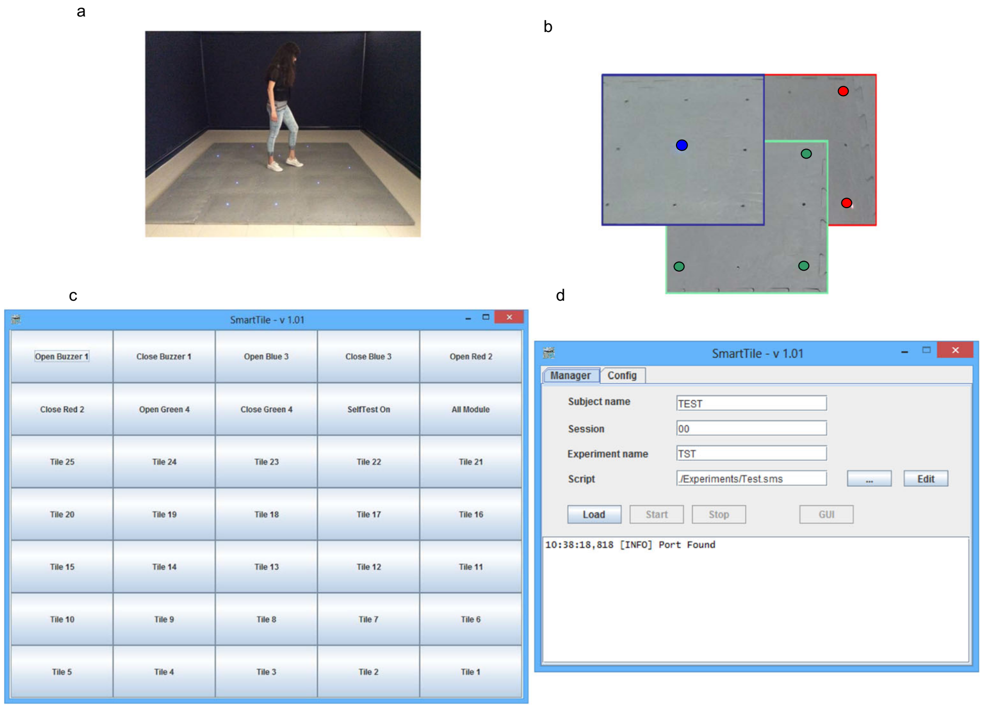

Fig. 1 SmarTile platform. (a) Picture of the platform (250 cm × 250 cm). (b) Different visual (blue, red, and green lights) feedback from the tiles. (c) The "graphical interface" of the platform allowing control of each tile independently. (d) The graphical interface of the platform to create or modify trials.

The end of each sequence was indicated by two consecutive short sounds.

- *Sequential Route B (SRB)* Visual stimuli were presented through blue lights that appeared on the platform. The lights turned on one by one (each tile was on for 2 s). Turning on a light stimulus caused the shutdown of the previous light. Then the subject heard a sound (lasting 2 s) that indicated he or she should stand on the platform and repeat the same sequence observed, walking on the tiles in the order they had previously been lit. Sequences of increasing length (from three to nine tiles) were presented until the subject failed to reproduce two out of three trials of a given length.

The end of each sequence was indicated by two short and consecutive sounds.

- *Simultaneous walking test (SWT)* Blue lights were lit simultaneously on multiple tiles in different spatial locations on the platform, forming an abstract figure (Fig. 2b). After the lights had been shut off, the subject heard a sound that indicated he or she should stand on the platform and recall the exact location of each tile by walking on the tiles previously lit and stopping on each tile, saying "this was on." Hence, the order of the tiles was not crucial in performing the task. Trials of increasing length (from three to nine tiles) were presented until the subject failed to reproduce two out of three trials of a given length. In this task, no sound indicated the end of the session, and the program was stopped by the experimenter after the last response of the subject on the platform.

At the beginning of each session, a sound indicated to the subject that something was going to occur on the platform. At the end of each presentation, another sound indicated that the subject had to step onto the platform and carry out the task, repeating what he or she had seen.

Reaching space In reaching space, VSWM was investigated by means of three tasks:

- *Corsi block-tapping test (CBT; Corsi, 1972)* The apparatus was composed of nine wooden blocks fixed on a board in a scattered array and numbered only on the experimenter's side. The experimenter tapped a sequence of blocks at the rate of one block per second, and the subject had to reproduce the same sequence in the same order, in the forward condition (CBT_F), or in the reverse order, in the backward condition (CBT_B). Sequences of increasing length (i.e., from a three-block sequence to a nine-block sequence) were presented until the subject failed to reproduce two out of three trials of a given length.
- *Sequential paper test (SqPT)* The apparatus was composed of a white sheet with a matrix of 25 cells, resembling the structure of the platform used in Experiment 1 (Fig. 3a). As in the CBT, the experimenter tapped a

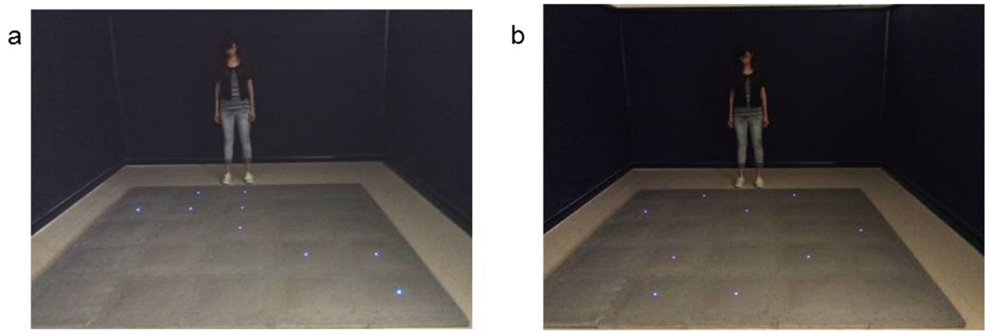

Fig. 2 Examples of sequential and simultaneous navigational tasks. (a) *Sequential Route A*: Example trial. The cues near each other light up, to draw a path that stays illuminated until the end of a specific sequence. (b) *Simultaneous walking test*: Example trial. All tiles turn on simultaneously, scattered in different parts of the platform, to form an abstract figure.

sequence of cells at the rate of one block per second, and the subject had to reproduce the same sequence in the same order (Fig. 3b).

Sequences of increasing length (from three to nine cells) were presented until the subject failed to reproduce two out of three trials of a given length.

- *Simultaneous paper test (SmPT)* The apparatus was the same as in the SqPT. Some of these cells were blackened (Fig. 3c), and the subject had to memorize the positions of the blackened squares. After a few seconds (1 s per blackened square), the experimenter covered the marked sheet of paper, and the subject was required to indicate on an empty array the exact locations of each blackened square. Hence, the order of the cells was not a component of this task. Trials of increasing length (from three to nine cells) were presented until the subject failed to reproduce two out of three trials of a given length.

Data analyses

In both the reaching and navigational tasks, subjects' performance was assessed via their span scores, corresponding to the longest sequence that they had correctly reproduced in each task. To ensure the normality of the data distribution, the span scores were expressed as proportions of the maximum achievable span and angular-transformed. Since two versions of the tasks were used (each one delivering a different set of sequences), the transformed data were first analyzed in a

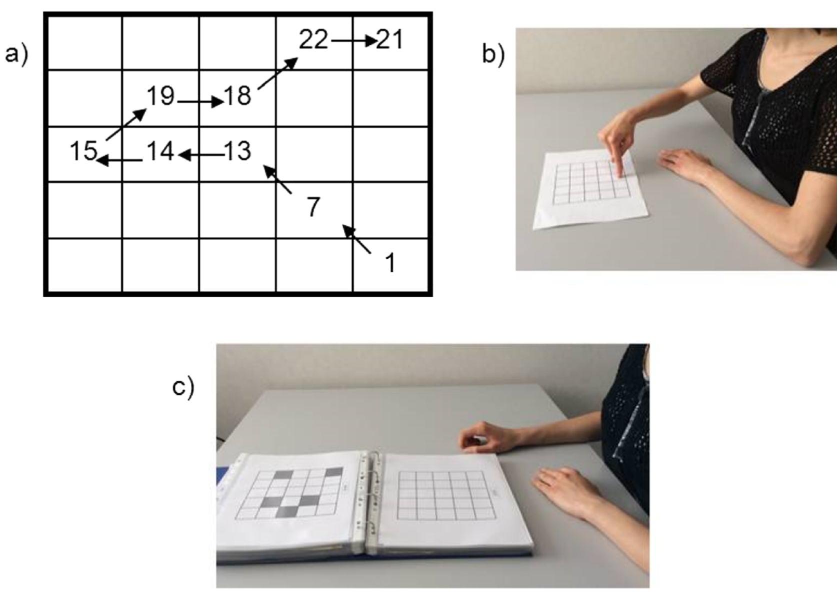

Fig. 3 Examples of sequential and simultaneous paper tasks. *Sequential paper test*: (a) Example of a sequence of cells tapped by the experimenter, at the rate of one block per second. (b) The white sheet with a matrix, on which the subject must indicate the cells previously touched by experimenter. (c) *Simultaneous paper test*: On the left side of the image is an example of the white sheet with a matrix of 25 cells where some of the cells are blackened (this sheet was covered before the subjects performed the task); on the right side of the image is the white sheet with a matrix on which the subject must indicate the cells that were previously blackened.

two-way analysis of variance [ANOVA] to check for any effect of the specific sequence used, using version (two levels, independent measures) and task (CBT, SqPT, SmPT, SRA, SRB, and SWT, repeated measures) as factors.

To investigate the factor structure underlying the tasks, the transformed data were submitted to both nonhierarchical and hierarchical factor analysis.

Results

Comparison between the two versions of the tests

The mean span scores obtained by the subjects in each task were above the cutoff typically used for the evaluation of the span (cutoff: <5) (see Table 1). This result indicates the absence of specific VSWM problems in the sample population, considering both space and presentation modality.

The results of the ANOVA on the span scores showed neither a main effect of version [$F(1, 69) = 1.40$, $p = .24$] nor an interaction between task and version [$F(5, 345) = 0.70$, $p = .57$] (Fig. 4). The absence of significant differences and interactions demonstrates that the two versions of the tasks are comparable and thus can be considered in parallel.

Instead, a significant main effect of task emerged [$F(5, 345) = 8.750$, $p < .001$]. The Newman–Keuls post hoc test of the task main effect showed that subjects had significantly better performance on the SmPT than on all the other tasks (*p*s < .01 in all the cases), better performance on the SRA task than on the SqPT ($p = .01$), and better performance on the SWT than on the SqPT ($p = .03$) (for the score range for each task, see Table 1). The subjects showed better performance in recovering the information presented in a simultaneous modality, independently of its presence in reaching or navigational space. This result suggests a significant effect of the type of coding used by the subjects to memorize the simultaneous stimuli versus the sequential ones.

Dimensionality of the tasks

A preliminary analysis of Mardia's (1970) multivariate asymmetry showed acceptable values for skewness (14.407, $p > .05$), whereas the coefficient for kurtosis was 61.56 ($p < .01$). This value suggests the presence of multivariate nonnormality in the distribution of the span scores for the present sample. Consequently, we performed exploratory factor analyses (principal axis factoring, promax rotation with kappa = 4) on the polychoric correlation matrix and on a Pearson's correlation matrix. Because the results from the analyses of the polychoric correlations and of the Pearson's correlations yielded the same results, only the latter will be presented here.

The Kaiser–Meyer–Olkin measure of sampling adequacy (.83) and Bartlett's test of sphericity [$\chi^2(15) = 120.84$, $p < .001$] supported the factorization of the correlation matrix of the six tasks. Because the scree test suggested a three-factor solution, accounting for 77.3% of the common variance (Table 2), three factors were extracted through the principal axis method.

After promax rotation (kappa = 4), the analysis showed a first factor accounting for 39.5% of the common variance that was saturated by the CBT, the SqPT, and (partially) the SRB task, suggesting that this factor underlies memory for sequences in the reaching space. Interestingly, in the SRB task, the presentation of the sequences was exactly the same as in the CBT and SqPT tasks; namely, each tile was turned off before the next tile in the sequence was turned on. The second factor, accounting for 35.9% of the common variance, was saturated in the SRA and SRB tasks, suggesting that this factor underlies memory for sequences in the navigational space. The third factor, accounting for 36.3% of the common variance, was saturated in the SmPT and the SWT, suggesting that this factor underlies memory for visual patterns, independent of whether they are presented in reaching or navigational space (Table 3).

The extraction of these three different factors further highlights the existence of important variables in the study of VSWM, such as the different effects of presentation modality (simultaneous and sequential) and of the analyzed space (reaching and navigational).

Moreover, it should be noted that the three factors were highly correlated with one another. Namely, the first factor was positively correlated with both the second factor (.746) and the third factor (.803), and the second factor was positively correlated with the third factor (.605). These correlations are hardly surprising, since all the tasks actually measure components of VSWM. However, the high correlation among the factors suggests that a higher-order (general) factor might be present. To investigate this hypothesis, the Pearson's correlation matrix was subjected to a Schmid–Leiman hierarchical factor analysis. In this analysis, higher-order factors are extracted that account for as much as possible of the correlation among the observed variables, whereas the lower-order, primary factors are residual factors that are not correlated with each other or with the higher-order factors. Consequently,

Table 1 Performance of 71 volunteers in each task

Reaching Space Test	Raw Score	Navigational Space Test	Raw Score
CBT	6.41 (1.15)	SRA	6.66 (1.23)
SqPT	5.97 (1.06)	SRB	6.15 (1.32)
SmPT	6.97 (1.76)	SWT	6.31 (1.76)

CBT = Corsi block-tapping test; SqPT = sequential paper test; SmPT = simultaneous paper test; SRA = Sequential Route A; SRB = Sequential Route B; SWT = simultaneous walking test. The data are reported as mean and standard deviation (*SD*).

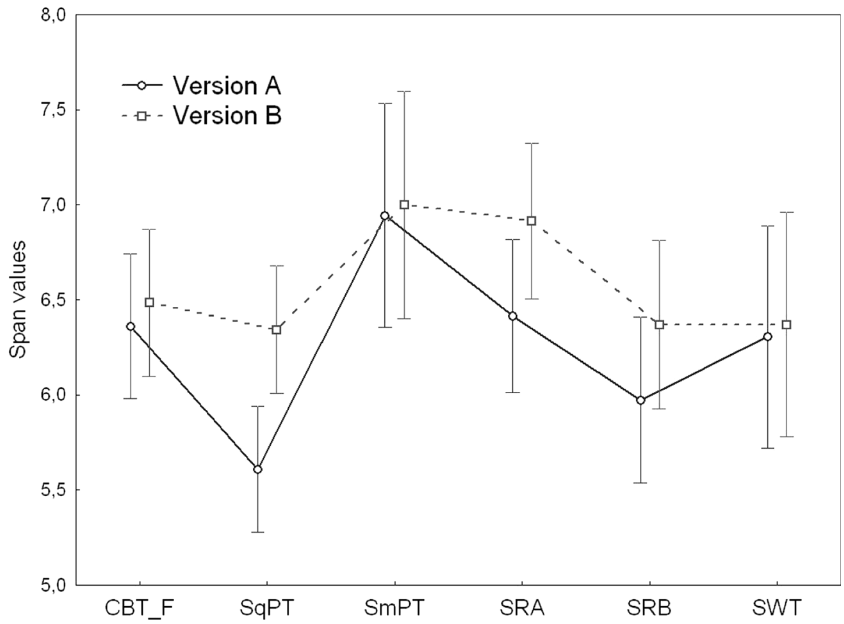

Fig. 4 Performance comparison between Version A and Version B. CBT_F = Corsi block-tapping test, forward version; SqPT = sequential paper test; SmPT = simultaneous paper test; SRA = Sequential Route A; SRB = Sequential Route B; SWT = simultaneous walking test.

each obtained factor represents the independent contribution of the factor in question (Schmid & Leiman, 1957).

It is worth noting that it is assumed that a higher-order factor emerges from a hierarchical factor analysis only if a general factor is truly latent in the analyzed correlation matrix. In the context of the present analysis, the higher-order factor would represent the shared variance among all the span measures, whereas the first-order factors would reflect the specific nature of each measure.

The results of the Schmid–Leiman hierarchical factor analysis showed that a general factor was indeed present and was saturated in all the investigated span measures, though its relevance to the CBT span scores was slightly weaker. With regard to the primary factors, the results confirm and clarify that the pattern emerged from the standard exploratory factor analysis reported above. Namely, two separate primary factors underlie memory for sequences in reaching space and in navigational space, whereas one single primary factor underlies memory for visual patterns, independent of whether they are presented in the reaching space or the navigational space (Table 4).

Because of the results of the exploratory factor analyses (both standard and hierarchical), the question arose as to why tests of memory for visual patterns do not differentiate between reaching and navigational spaces. One possible explanation calls into account how individuals explore the visual input. That is, it is possible that individuals follow with their gaze the different spatial locations when those locations are presented to them in a sequence. Instead, when visual patterns are presented, individuals do not move their gaze from one location to another, but rather fixate on one point in space and code the visual pattern as a single configuration. Such a strategy would be effective independent of the space wherein the

Table 2 Eigenvalues and percentages of explained variance

Eigenvalue	% of Variance	Cumulative%
3.100	51.660	51.660
0.810	13.508	65.169
0.728	12.131	77.300
0.541	9.019	86.319
0.446	7.429	93.748
0.375	6.252	100.000

Scree test results: three-factor solution extracted through the principal axis method, accounting for 77.3% of the common variance.

Table 3 Rotated factor matrix resulting from the factor analysis (principal axis, promax rotation) on the span scores (angular transformed) of the different tasks used

	Factor 1	Factor 2	Factor 3
CBT	**.598**	– .082	– .060
SqPT	**.658**	.012	.068
SmPT	.090	.283	**.409**
SRA	– .089	**.964**	– .085
SRB	**.325**	**.379**	.144
SWT	– .041	– .099	**.909**
Prop. expl. variance	.396	.360	.363

CBT = Corsi block-tapping test; SqPT = sequential paper test; SmPT = simultaneous paper test; SRA = Sequential Route A; SRB = Sequential Route B; SWT = simultaneous walking test.

Table 4 Results of the Schmid–Leiman hierarchical factor analysis

	Higher-Order Factor	F1	F2	F3
CBT	.434	**.191**	– .018	– .001
SqPT	.687	**.253**	.007	.021
SmPT	.683	– .056	.140	**.245**
SRA	.605	– .042	**.473**	– .101
SRB	.744	.089	**.240**	.022
SWT	.648	.051	– .134	**.353**

CBT = Corsi block-tapping test; SqPT = sequential paper test; SmPT = simultaneous paper test; SRA = Sequential Route A; SRB = Sequential Route B; SWT = simultaneous walking test.

pattern is presented. If this is true, then the dimensionality of the tasks would not point to different memory processes based on the space (navigational and reaching) and stimulus (sequences and pattern), but would merely depend on the individual modality adopted to cope with the task. To investigate this hypothesis, we collected both performance and eye movement data from a new group of subjects who were subjected to all the tests described above, except for the CBT and SqPT.

Experiment 2

Experiment 2 was a pilot study that aimed to disentangle spatial sequential and simultaneous processes based on the hypothesis that during simultaneous presentation, items that are located in different positions can each be scanned by the subject several times, whereas in a sequential presentation, the order of encoding is imposed, and the rescanning of a single position is less likely (Pazzaglia & Cornoldi, 1999). To address this issue, we collected eye movement data from a new group of subjects in the same sequential and simultaneous tests as in Experiment 1. Our prediction was that simultaneous presentation would improve the subjects' performances.

Method

Subjects

Ten healthy subjects [M/F: 4/6; mean age: 25.80 (*SD* = 2.15); mean years of education: 16.80 (*SD* = 1.03)] participated in the experiment. All the subjects had normal or corrected-to-normal sight.

All the subjects were recruited from the Department of Psychology, Sapienza University of Rome. The experimental procedures were approved by the Institutional Review Board of the Department of Psychology. The study has been carried out in accordance with the Code of Ethics of the World Medical Association (Declaration of Helsinki). Written consent was obtained from each subject.

Procedures

The general procedure was the same as in Experiment 1, except that eye movements were recorded during the presentation of the to-be-remembered stimuli. The SRA, SRB, SmPT, and SWT were administered using the same procedures as described above. Only trials with lengths of three, six, and eight tiles were presented. The CBT and SqPT were not administered, because in order to register the eye tracking, a computerized version should have been used instead of the original version in which the experimenter presented the sequences using their finger, and this difference in procedure might influence the performances. Indeed, recent evidence by Tedesco et al. (2017) showed that when a clinical population is presented with two similar tasks—such as the "walking Corsi," with an examiner that walks on the carpet, and the "magic carpet," in which a computer lights up the tiles in the sequence—different mental processes are implicated in solving the tasks. The authors suggested that in the walking Corsi test, the experimenter who performs the sequence on the carpet allows the patient to simulate the action mentally, whereas this simulation cannot be triggered by the magic carpet (Tedesco et al., 2017). These data led us to exclude the possibility of comparing computer versions of the CBT and SqPT to our original tasks; thus, we did not include these two tasks in Experiment 2.

Eye movement recording

During the SRA, SRB, and SWT, eye movements were collected using Tobii infrared eyetracking glasses (Version 1, Tobii, Sweden), composed of a head-mounted monocular (right eye only) system resembling a pair of regular glasses. The sampling frequency was 30 Hz, and the recording visual angle was 56° × 40°. The data were collected on a mini-SD card through the Recording Assistant.

During the SmPT task, eye movements were recorded using the infrared Tobii Eye-Tracking X50 system. The sampling frequency was 50 Hz, stimuli were presented on a 17-in. monitor, and subjects were seated at a distance of 60 cm.

The use of two different eye-tracking systems was based on the different purposes for their use. Indeed, for observations of two-dimensional structures, a stationary eye tracker is more practical and precise, while a head-mounted eye-tracking system is especially helpful when the object to be observed has a three-dimensional structure or when respondents need to be able to move freely, as in this case.

Both recording systems were calibrated for each individual.

Data analyses

For each subject and task, the proportions of times the tiles not belonging to the presented stimulus were fixated on (*p_out*) were computed and angular transformed. They were separately analyzed in a repeated measures two-way ANOVA with task (SRA, SRB, SWT, and SmPT) and length (three, six, and eight tiles) as factors.

Results

The results of the analysis on the (*p_out*) data showed a significant main effect of task [$F(3, 27) = 24.58$, $p < .001$], whereas neither the main effect of length nor the task by length interaction was significant [$F(2, 18) = 0.86$, $p = .44$, and $F(6, 54) = 1.77$, $p = .12$, respectively]. The Newman–Keuls post hoc test of the task main effect showed that subjects fixated on tiles not belonging to the presented stimulus significantly (*p*s < .03 in all the cases) fewer times when performing the sequential tasks in the navigational space (SRA and SRB) than when performing the simultaneous tasks in the navigational and reaching spaces (SWT and SmPT), whereas no significant differences emerged between the SRA and SRB tasks or between the SWT and SMPT (Fig. 5). These data demonstrate that subjects use different eye fixation strategies according to the presentation modality (sequential or simultaneous) of the stimuli and independently of reaching or navigational space.

Discussion

In the present study, we aimed to gain some further insights into characteristics and functioning of VSWM by using an integrated neuropsychological and neurophysiological approach.

The first aim of the study was to develop a battery of tests and a new tool able to predict, assess and respond to challenges for VSWM abilities, considering the multifarious components of this function. To fulfill this purpose, we have examined the psychometric properties and the factor structure of the tasks administered by means of the SMARTile platform and of specific paper-based tasks.

In accordance with the theoretical proposition of differences in VSWM related to the spatial condition (reaching or navigational) and to the modality of item presentation (sequentially or simultaneously), in Experiment 1, analysis using the principal axis method extracted three factors, demonstrating the presence of a distinct memory for sequences in the reaching space, separate from sequences in the navigational space; the same distinction between reaching and navigational space has not been found in memory for visual patterns that rely on simultaneous presentation. We hypothesized that this finding might be related to a different modality of eye gaze exploration that the individuals implemented according to pattern or sequence presentation (see Exp. 2).

Previous studies have demonstrated that the system underlying route memory is dissociated from the system that memorizes sequences of locations in near space, as shown by clinical observations of patients who failed to store and recall sequences on the CBT and were never affected by navigational disorders, and by observations of patients affected by topographical disorientation who perform flawlessly on the Corsi test (Palermo et al., 2014; Mammarella et al., 2009). Conversely, for the simultaneous modality, the effect of near space versus navigational space is less clear. Indeed, in clinical populations, studies investigated the effect of simultaneous

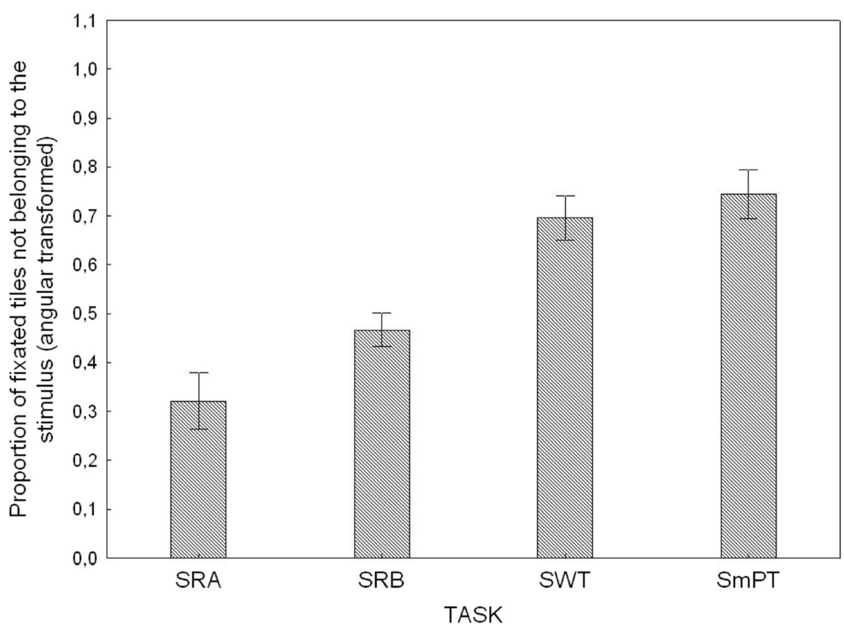

Fig. 5 Mean proportions of fixated tiles not belonging to the presented stimulus (angular-transformed). The standard error is reported. SRA = Sequential Route A; SRB = Sequential Route B; SWT = simultaneous walking test; SmPT = simultaneous paper test.

presentation of stimuli on WM performance only in reaching space, thus not allowing comparison of the performances between reaching and navigational spaces.

The effects of sequential or simultaneous presentation of stimuli on performance have been recently described in children with a nonverbal (visuospatial) learning disability (NLD) and in patients with unilateral neglect (Mammarella et al., 2009; Wansard et al., 2015), showing a dissociation according to sequential and simultaneous presentation modality. Specifically, whereas patients affected by unilateral neglect performed worse when the stimuli were presented sequentially than when they were presented simultaneously, children with NLD showed the opposite behavior (Mammarella et al., 2009; Wansard et al., 2015).

Nevertheless, no previous study investigated all aspects of VSWM in the same population of subjects considering both reaching and navigational space and sequential and simultaneous stimulus presentation. Therefore, in the present study, we assessed the VSWM profile of healthy individuals, taking into account different conditions of space (reaching or navigational) and stimulus presentation (sequential or simultaneous). We demonstrated that in both reaching and navigational spaces, the best performances occurred with the simultaneous presentation modality.

In healthy subjects, better performance when the locations to be remembered were presented simultaneously rather than sequentially was also reported by Lecerf and De Ribaupierre (2005), who hypothesized that simultaneous presentation should encourage using both intrafigural encoding (i.e., making relations between different positions independently from an external frame) and extrafigural encoding (which requires the encoding of each position separately from one another and relative to an external frame of reference), leading to better performance. In contrast, sequential presentation makes the creation of patterns much more complex than in the simultaneous condition. According to the authors, due to the unpredictability of the positions, sequential presentation would require more time. Indeed, simultaneous presentation allows better allocation of attentional resources and more efficient rehearsal because the subjects can decide in which order the locations will be encoded, and they encode them essentially at their own pace and rescan some or all of them (Lecerf & De Ribaupierre, 2005).

Taking into account this theoretical framework, we used eye-tracking monitoring to better understand the effect of presentation modality in a second group of healthy subjects (Exp. 2). The results showed that, as compared to the sequential conditions, in the simultaneous tasks subjects fixated on significantly fewer tiles among the presented stimuli. We hypothesize that this eye fixation strategy improves performance because of the possibility to fixate on one point in space and code the visual pattern as a single configuration. According to these results, the effects of the tasks on performance, as emerged from the factor analysis, seem to be actually due to better allocation of attentional resources of working memory processes and to more efficient rehearsal with no differentiation between navigational and reaching spaces when coding simultaneous stimuli. Thus, the difference between simultaneous and sequential presentation could be due to a specific strategy used by subjects to explore stimuli grouped in space (simultaneous or pattern stimuli).

In summary, the present study showed a complete view of healthy individuals' VSWM performances, presenting a new battery of tests and tools able to accurately investigate the subcomponents of this function.

Indeed, analysis using the principal axis method allowed us to demonstrate that the SMARTile platform provides an accurate assessment of the overall aspects of VSWM function in navigational space and that the paper tests used in this study effectively measure the same components in reaching space, thereby allowing the generation of specific and well-defined VSWM profiles.

However, it is worth noting that there are some limitations related to the intrinsic characteristics of this new protocol. Indeed, it can be performed only by subjects who are able to ambulate independently and who do not show severe impairment in eye movements.

The use of specific motor scales that quantify the patients' clinical profiles will help clinicians to select the patients suitable for testing and will provide motor scores to correlate with the behavioral data in order to assess the possible effects of motor impairment on performance on these tasks.

Despite these limits, our protocol could be highly appropriate for specific diseases that do not cause serious difficulties in walking and that are known to affect specific components of visuospatial function.

Indeed, our battery could be a useful instrument in pathologies for which topographical orientation impairments are reported, as in developmental topographical disorientation pathology (Bianchini et al., 2010; Bianchini, Palermo, et al., 2014; Iaria, Bogod, Fox, & Barton, 2009; Palermo, Foti, Ferlazzo, Guariglia, & Petrosini, 2013; Palermo et al., 2014), mild cognitive impairment (Rusconi, Suardi, Zanetti, & Rozzini, 2015), early stages of Alzheimer's disease (Bianchini, Di Vita, et al., 2014), and Williams syndrome (Lanfranchi, De Mori, Mammarella, Carretti, & Vianello, 2015; Nardini, Atkinson, Braddick, & Burgess, 2008).

Another point that needs to be addressed is whether the presence/absence of the examiner influences the learning process.

Indeed, in the CBT and SqPT, the subject saw the examiner performing the task, whereas in all navigational tasks and in the SmPT, the presentation was based on the computerized activation of the tiles or the simultaneous presentation of the stimuli on a sheet and, therefore, was not linked to the action of the examiner. Taking this into account, we cannot exclude the possible involvement of the mirror system and of an imitation learning process (Buccino et al., 2004) when the subjects stored the CBT and SqPT sequences. However, an effect of the presence/absence of the examiner was not supported by the factor analysis, which demonstrated that the 1st factor (which includes the sequential tasks, CBT and SqPT, in reaching space) was partially saturated also in the navigational SRB task. It is important to emphasize that in the SRB task, the sequences are presented in a modality more similar to that of the CBT and SqPT, but without the examiner. This information allows the supposition that the effect of the modality of presentation (sequential or simultaneous) overrides the effect of the space (reaching or navigational) or the examiner (present/absent), further demonstrating the efficacy of the developed neuropsychological battery in testing the visuospatial coding abilities.

Finally, although it was not the main purpose of the work, it might be interesting to distinguish VSWM performance with regard to gender and age. Further studies will be needed to test the effects of gender and age in this neuropsychological battery.

In conclusion, given that clinical studies have shown the presence of different patterns of VSWM alterations in different neurological populations (Aguirre & D'Esposito, 1999; Farrell, 1996), it is important to characterize the profile of performances in various neurological patients using specific tests that allow for accurate differentiation between the components of VSWM. From this perspective, SMARTile and the neuropsychological battery set up in the present study may provide an instrument that allows the detection of specific and partial deficits, proving extremely valuable in clinical practice.

Author note The editing support of American Journal Experts is acknowledged. The authors have no conflicts of interest to declare. No targeted funding reported.

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