Contextual reminders fail to trigger memory reconsolidation in aged rats and aged humans

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ABSTRACT

There is strong evidence that hippocampal memory returns to a labile state upon reactivation, initiating a reconsolidation process that restabilizes it and allows for its updating. Normal aging is associated with deficits in episodic memory processes. However, the effects of aging on memory reconsolidation and its neural substrate remain largely unknown, and an animal model is lacking. In this study we investigated the effects of aging on context-dependent reconsolidation using an episodic set-learning task in humans and an analogous set-learning spatial task in rats. In both tasks, young and older subjects learned a set of objects (humans) or feeder locations (rats; Set 1) in Context A on Day 1. On Day 2, a different set (Set 2) was learned in either Context A (Reminder condition) or Context B (No Reminder condition). On Day 3, subjects were instructed (humans) or cued (rats) to recall Set 1. Young rats and humans in the Reminder condition falsely recalled significantly more items from Set 2 than those in the No Reminder condition, suggesting that the reminder context triggered a reactivation of Set 1 on Day 2 and allowed the integration of Set 2 items into Set 1. In both species, older subjects displayed a different pattern of results than young subjects. In aged rats, there was no difference between conditions in the level of falsely recalled Set 2 items (intrusions). Older humans in the No Reminder condition made significantly more intrusions than those in the Reminder condition. Follow-up control experiments in aged rats suggested that intrusions in older animals reflected general interference, independent of context manipulations. We conclude that contextual reminders are not sufficient to trigger memory updating in aged rats or aged humans, unlike in younger individuals. Future studies using this animal model should further our understanding of the role of the hippocampus in memory maintenance and updating during normal aging.

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1. Introduction

Consolidated memories do not remain stable but return to a labile state upon reactivation or retrieval, requiring a "reconsolidation" process in order to once again become resistant to interference. Because reconsolidation occurs across a broad range of species and memory paradigms, it appears to be a fundamental stage of memory processing (Jones, Bukoski, Nadel, & Fellous, 2012; Nader & Einarsson, 2010; Sara, 2000; Tronson & Taylor, 2007).

At the behavioral level, memory reactivation leads to updating when new information is present at the time of reactivation. Hupbach, Gomez, Hardt, and Nadel (2007), Hupbach, Hardt, Gomez, and Nadel (2008) found that young adults who learned a new set of objects in the same spatial context as a previously learned set falsely recalled items from the second set (Set 2 intrusions) when asked to recall the first, whereas subjects who learned the sets in different contexts did not. The effect was not attributable to source memory error (Hupbach, Gomez, & Nadel, 2009). Therefore, it appears that the context reactivated the memory for the first set, returning it to a labile state and allowing for new items to become incorporated into the memory (Nadel, 2008).

We adapted the Hupbach, Hardt, Gomez, and Nadel (2008) paradigm in order to investigate updating of appetitively-motivated spatial memories in young adult rats (Jones et al., 2012). In this study, animals learned on Day 1 to obtain sugar water rewards from 3 feeders (Set 1) on an open-field arena. On Day 2, the rats were trained to

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run to 3 different feeders (Set 2) in either the same (Reminder condition) or a different (No Reminder condition) experimental context than that used on Day 1. On Day 3, they were cued to recall Set 1 feeders. Consistent with the human study, rats in the Reminder condition made significantly more visits to Set 2 feeders (intrusions) during Set 1 recall than rats in the No Reminder condition, suggesting that the reminder triggered reconsolidation and allowed for the integration of some of the Set 2 items into Set 1.

Aging is associated with episodic memory impairments in humans (Rajah, Kromas, Han, & Pruessner, 2010) and other mammals (Burke & Barnes, 2006). These impairments have been linked to deficits in hippocampal synaptic plasticity and changes in hippocampal network activity associated with memory consolidation. It is possible that aging leads to changes in the reconsolidation process as well. However, to our knowledge, the effects of aging on reconsolidation have not been investigated.

Using reminder-dependent memory updating as a behavioral marker of reconsolidation, the current study examines the effects of aging on contextually-triggered reconsolidation of spatial memories in rats and episodic memories in humans. If reconsolidation is triggered and occurs similarly across ages, then we expect to replicate in older animals the previous results seen in young humans and rats by measuring more intrusions in the Reminder group than in the No Reminder group. However, if the reconsolidation process is altered in aged subjects, then we expect there to be no difference in the amount of intrusions expressed in the Reminder and No Reminder groups.

2. Materials and methods

2.1. Animal experiments

Parts of these methods have been described previously in Jones et al. (2012). The animal procedures are specifically designed to be as close as possible to the human experiments so comparisons can be made across species.

2.1.1. Animals

Eight adult and 16 aged male Fischer 344 rats were obtained from the National Institute on Aging for this study. Adult rats were obtained at 8–12 months of age and completed the experimental procedure (including pretraining) within ~2 months of arrival. Aged rats were obtained at 22 months, started experiments (after pretraining) at 24 months, and completed experiments by 26 months of age (see Table S1 for experimental timeline). All rats were maintained on a reversed 24 h light/dark cycle, and food restricted to 85% of their *ad libitum* weight for the duration of the study. Experiments were conducted in dim light conditions, in the dark (active) phase. Rats were used across multiple experiments and conditions, with the order of conditions counterbalanced for each experiment (see Table S2 and supplemental methods for experiment order). The average time between successive experiments was ~4 days (SD = 3.6), and the average time between 1st and last experiments was ~48 days (SD = 11.9). While all of the rats were reused, not all were available for the entire study (e.g. due to declining health and mortality in aged animals), resulting in various numbers of animals across different experiments and conditions (as noted in the figures). Different feeder sets and different combinations of sensory cues (contexts) were used for each different condition and experiment. In order to ensure that the animals did not have a preference for sets from previous experiments, time between experiments always included at least 1 day of random training, during which rats were repeatedly cued to visit all eight feeders in random order. All procedures were in accordance with the animal care guidelines of the University of Arizona and approved by the IACUC.

2.1.2. Apparatus

The apparatus used was a 5′ wide open-field circular arena which contained 8 evenly spaced feeders around the edge (Bower, Euston, & McNaughton, 2005; Jones et al., 2012). The arena was lined by a 1ft tall wall at its periphery. Attached to each feeder was a LED light. Reward was a drop of sugar water (0.2 g/mL). LED onset could be delayed to assess whether rats went to the feeders using memory alone. Rats were tracked by an overhead camera, and the feeders and lights were automatically controlled by a computer and custom written software (Labview, National Instrument). Spatial context was manipulated as a combination of an odor, a floor texture (e.g. carpet), and 2–3 distal visual cues attached to curtains surrounding the arena. A local cue was kept constant at the edge of the arena to promote recognition of the feeder arrangement across days (i.e. so that rats could orient themselves on the table despite contextual changes).

2.1.3. Procedure

2.1.3.1. Experiment 1.1 – the role of context as a reminder. All animals were pretrained to randomly go to a blinking light to get a reward at the adjacent feeder. On Day 1, rats were cued by blinking lights to run to 3 pseudo-randomly presented feeders (learning phase, e.g. Set 1 = 3, 5, 7) in the context of specific floor texture, odor, and distal visual cues (Context A, Fig. 1A). After young rats visited each feeder 50 times (150 rewards), the light cues were delayed by 15 s, and the rats had to choose the feeders from memory (test phase). The 3 feeders were baited in pseudorandom order such that the same feeder could not be rewarded twice in a row and each of the 3 feeders must be visited before the next triplet of feeders could be baited. Rats could visit the correct feeders in any order but would receive reward only at the baited feeder (thus, they had a 50% chance of reward when visiting a correct feeder after having visited a different correct feeder, and a 33% chance of reward after having visited an incorrect feeder). This ensured visitation to all 3 correct feeders. The delay was reset if the rat reached the baited feeder within less than 15 s, and the next light cue was activated with a new delay of 15 s. The procedure was the same for aged rats except, due to their reduced velocity and endurance, they visited each feeder 25 times (75 rewards), and the light cues were delayed by 20 s. For all rats, the criterion was reached when they visited 15 correct feeders consecutively with no more than 2 feeders being cued (i.e. the rats beat the light at least 13 out of 15 times).

On Day 2, the procedure was the same as for Day 1 except that a different set of feeders (e.g. Set 2 = 1, 4, 6) was used. There were two conditions on Day 2. In the Reminder condition, rats learned Set 2 in Context A. There was no explicit recall of Set 1 on Day 2, so that Context A was the only reminder of Set 1. In the No Reminder condition, rats learned Set 2 in the context of different floor texture, odor, and distal visual cues (Context B). On Day 3, the rats were cued to recall Set 1 in Context A. They received light cues and rewards only at Set 1 feeders, as during the light-delay phase of training. The first light cue was immediate in order to cue the rats to the correct set of feeders to recall, but the subsequent cues were delayed by 20 s each (15 s for young rats). The test continued until performance reached the same criterion as during training.

2.1.3.2. Experiment 1.2 – the selectivity of the reminder effect. Only aged rats performed Experiment 1.2. The training and test procedures for Experiment 1.2 were exactly the same as for Experiment 1.1 except that the rats were cued to recall Set 2 rather than Set 1 on Day 3 (Fig. 1B).

2.1.3.3. Experiment 1.3 – the time course of the reminder effect. Only aged rats performed Experiment 1.3. The training and test procedures for Experiment 1.3 were exactly the same as for Experiment

A   Experiment 1.1   **B**   Experiment 1.2
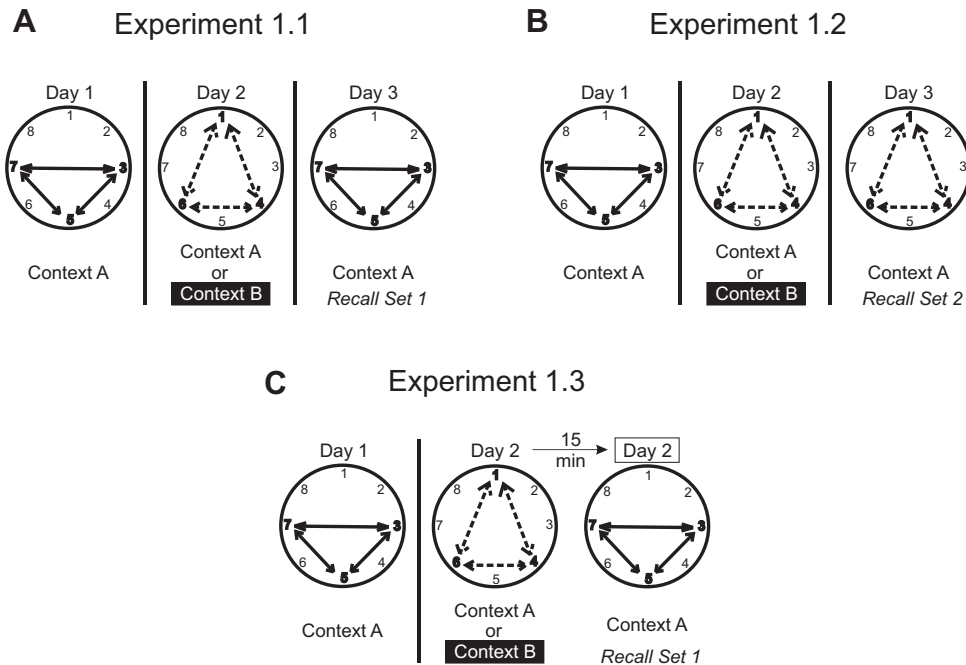
C   Experiment 1.3

Fig. 1. Designs of rat experiments. (A) Experiment 1.1. Rats learned Set 1 on Day 1 in Context A. Rats learned Set 2 on Day 2 in either Context A (Reminder condition) or Context B (No Reminder condition). Rats were cued to recall Set 1 in Context A on Day 3. B. Experiment 1.2. Same as Experiment 1.1, but rats were cued to recall Set 2 instead of Set 1 in Context A on Day 3. C. Experiment 1.3. Same as Experiment 1.1, except that rats were cued to recall Set 1 in Context A on Day 2 either 15 min or 4 h following training for Set 2. figure adapted from Jones et al. (2012).

1.1 except that the cued recall of Set 1 took place either 15 min or 4 h following Set 2 training on Day 2 rather than on Day 3 (Fig. 1C).

2.1.4. Data and statistical analysis

The feeder choices of the rat were recorded by both the experimenter and the computer program. Visits to each feeder type (Set 1, Set 2, and No Set: the 2 remaining feeders that were part of neither Set 1 nor Set 2) were expressed as a percentage of the total number of feeders visited. These values were normalized to account for the difference between the numbers of each feeder type. When analyzing recall, visits which were cued by the LEDs (when the rat failed to visit the correct feeder during the delay) were not included. To calculate the percent correct recall and intrusions for each rat, the fraction of No Set feeders was subtracted from the fraction of Set 1 and Set 2 feeders, respectively, in order to control for baseline errors. To calculate percent recall and intrusions in Experiment 1.2, the fraction of No Set feeders was subtracted from the fraction of Set 2 and Set 1 feeders, respectively. Data points more than 3 standard deviations from the mean were considered outliers and excluded from analysis.

Differences between groups were analyzed using either ANOVAs or two-tailed *t*-tests. Equality of variances was assessed with Levene's Test, and adjusted *p*-values are reported when applicable. Post hoc pair-wise comparisons were made using Tukey's HSD test or two-tailed *t*-tests (equal variances) or the Games–Howell Test (unequal variances). Significance levels were set to 0.05. All tests were performed using SigmaStat (SYSTAT, San Jose, California) or SPSS.

2.2. Human experiments

2.2.1. Participants

A group of 40 community dwelling adults over the age of 65 without previous neurological problems participated in the experiment. The participants were recruited from the subject pool from the Aging and Cognition Laboratory at the University of Arizona. All older adults had received neuropsychological testing within

the past two years[2] to assess their frontal and medial temporal lobe function, and have been assigned *z*-scores representing their performance relative to a normative group of 227 older adults on two composite measures. Tests included in the frontal composite (FL factor) thought to represent executive functions associated with working memory (Glisky & Kong, 2008; Glisky, Rubin, & Davidson, 2001) include the number of categories achieved on the modified Wisconsin Card Sorting Test (Hart, Kwentus, Wade, & Taylor, 1988), Mental Arithmetic from the Weschler Adult Intelligence Scale – Revised (WAIS-R; Wechsler, 1981), Mental Control from the Wechsler Memory Scale – III (WMS-III; Wechsler, 1997), Backward Digit Span from the WMS-III, and the total number of words generated on the Controlled Oral Word Association Test (Benton & Hamsher, 1976). Neuropsychological tests contributing to the medial temporal composite (MTL factor) thought to reflect retention and consolidation (Glisky & Kong, 2008; Glisky et al., 2001) include Logical Memory I, Verbal Paired Associates I, Faces I (all from WMS-III), Visual Paired Associates II (WMS-R, 1987), and Long-Delayed Cued Recall from the California Verbal Learning Test (Delis, Kramer, Kaplan, & Ober, 1987).

Participants were assigned to one of four groups (*N* = 10 per group) that were created by crossing the FL and MTL factors. The four groups were thus comprised of those individuals who were above average on both factors (HH), below average on both factors (LL), high on frontal/low on medial temporal (HL), and low on frontal/high on medial temporal (LH). Characteristics of each group are presented in Table S3. Separate 2 (High vs. Low FL) × 2 (High vs. Low MTL) analyses of variance (ANOVAs) were conducted to test for differences in age, education, and scores on the Mini-Mental Status Examination (MMSE; Folstein, Folstein, & McHugh, 1975) as a function of neuropsychological group. There were no significant differences in age or education between high and low FL groups or between high and low MTL groups. There were

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[2] Twenty-eight people were tested in the past year, 9 in the past 18 months, and 3 within 19–24 months. More recent tests on a subset of these individuals suggest that participants are unlikely to have changed neuropsychological categories since their original testing.

differences between high and low groups on MMSE scores, $F(1,36) = 5.77$, $p = .02$ (for both FL and MTL factors). Although older adults with above average function on the FL and MTL composite measures had slightly higher MMSE scores, all of the participants were within the normal range (i.e., $\geqslant 26$). A control group of 27 young adults (mean age = 19.8, SD = 2.8; mean education = 12.7, SD = 1.3) was also included.

2.2.2. Materials

Materials were identical to those used by Hupbach et al. (2008) except set lengths were reduced from 20 to 15 unrelated objects for the older adults. The two groups of objects were counterbalanced so that half of the participants in each group received Set 1 on the first day and the other half received Set 2 on the first day.

2.2.3. Procedure

The procedure also was similar to that used by Hupbach et al. (2008). Participants were informed that they would have to learn different sets of objects on different days. The three sessions took place on Monday, Wednesday, and Friday of the same week. On Day 1 the experimenter took the objects out of a box one at a time and placed them in a blue basket. The participants were asked to name each item as it was shown to them and to try to remember the objects so that they could recall them later. After all of the items were presented, they were put away by the experimenter and the participants were asked to recall as many items as they could, in any order. This procedure was repeated until the older adults could recall at least 13 of the 15 items on the set or until they had completed five learning trials. Younger adults continued until they recalled at least 17 of the 20 objects or completed four learning trials.

On Day 2, participants were pseudo-randomly assigned to one of two experimental conditions—Reminder or No Reminder, such that the two experimental groups did not differ on any of the demographic variables. People in the Reminder group went to the same room as Day 1 while those in the No Reminder group went to a different room. In both conditions, a different experimenter tested the participant on Day 2. Thus, as in previous studies, only the context differed between the Reminder and No Reminder groups. Participants in both groups were asked to learn a second set of objects. This procedure differed from Day 1 so that the task did not serve as a reminder. All of the objects in the second set were placed in front of the participants. They were asked to name the objects and were given 30 s to study them. The objects were then removed and participants were asked to recall them in any order. If they recalled less than 13 items (17 for young), the procedure was repeated until they could recall 13 (17) or more items or until they had reached five (four) trials.

On Day 3, all participants returned to the same room with the same experimenter as on Day 1 and were asked to recall as many objects as they could from Day 1. When participants indicated that they were unable to recall any more objects, they took a 30 s break during which the experimenter engaged the participants in a conversation about an unrelated topic. They were then asked to recall the items again. This was repeated for four recall trials. Average recall performance across the four trials served as the dependent measure. Participants were then given a brief questionnaire to see if they had noticed whether the two sets had been initially learned in the same or a different room and with the same or a different experimenter.

3. Results

3.1. Memory reconsolidation in adult and aged rats

The purpose of the rat study was to investigate the effects of aging on memory performance in a spatial reconsolidation task,

with the goal of comparing the results with that of the human experiments as in our previous study in young rats (Jones et al., 2012). The main experiment (1.1) assessed whether re-exposure to a spatial context triggers memory intrusions in aged animals. Experiment 1.2 was designed to measure the influence of set confusion on memory intrusions, and Experiment 1.3 investigated the time course of the intrusion effect. A supplemental experiment examined the effect of the retrieval context (see Supplemental Results and Fig. S2). Fig. 1 outlines the experimental designs (see Section 2 for details).
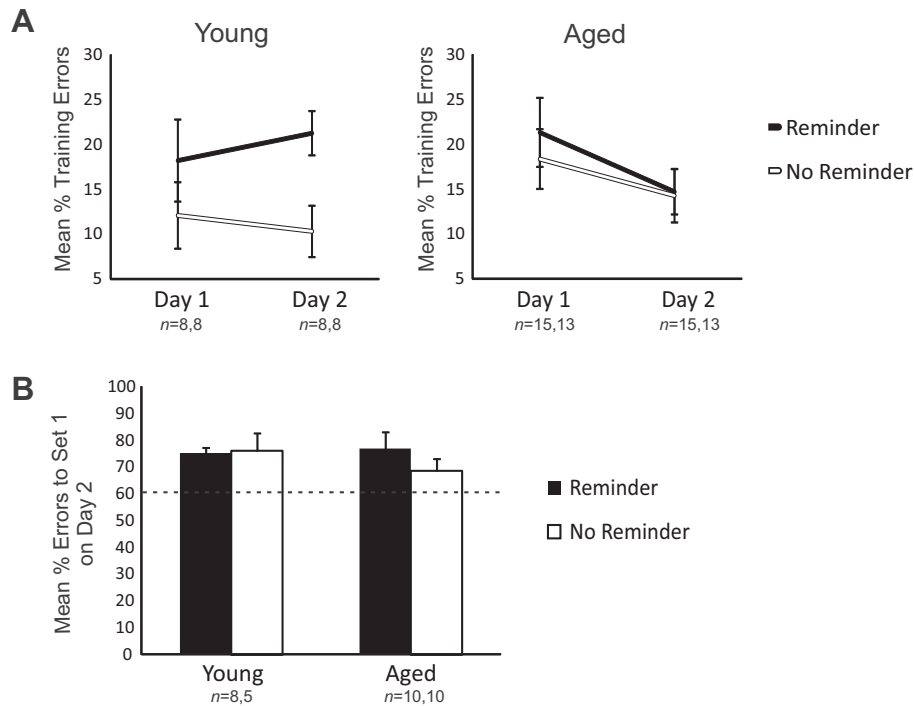
3.1.1. Learning performance on Days 1 and 2 in Experiment 1.1

Rats learned a spatial set of feeders (Set 1) on Day 1 and a different set of feeders (Set 2) on Day 2. The total number of feeder visits (including errors) required to reach the learning criterion during the test phase did not significantly differ between young and aged rats on either day (p's > 0.05). In order to analyze memory acquisition, the percent errors (visits to feeders not belonging to the current set) made during the test phase of the task were recorded on both days for each rat. Fig. 2A shows the learning performance plotted separately for the young and aged groups. A $2 \times 2 \times 2$ mixed-design ANOVA with group and condition as between-subjects factors and day as within-subjects factor showed no significant main effects and no significant interactions (all p's > 0.05) Because non-significant effects could be due to low statistical power, we more closely examined potential learning differences between the age groups by conducting separate 2 (group) $\times$ 2 (condition) ANOVAs for each day. There was no significant main effect or interaction on either day, though we note the non-significant effect of condition ($F(1,40) = 3.722$, $p = 0.061$) and interaction ($F(1,40) = 3.163$, $p = 0.083$) on Day 2 (all other p's > 0.25). A more fine-grained analysis of within-session learning performance (% correct feeders visited during each 10% increment of training) indicated that aged rats performed at least as well as young rats over the course of learning on each day (see Supplemental Results and Fig. S1). These results suggest that learning was equivalent in young and aged rats for both Set 1 and Set 2.

It is possible that aged rats learned equally as well as young rats but failed to consolidate the memory for Set 1. Reasoning that memory for Set 1 could manifest as erroneous visits to Set 1 feeders on Day 2, we assessed whether the proportion of these visits differed between age groups (Fig. 2B). The percentage of errors attributable to Set 1 was calculated for all rats that made at least 2 errors during the test phase of Set 2 training. One-sample t-tests indicated that both young ($t = 5.926$, $p < 0.001$) and aged ($t = 3.489$, $p = 0.001$) rats visited Set 1 feeders significantly more than chance (because sets were 3-feeders long and there were 8 possible feeders, the probability that rats would make errors to Set 1 feeders by chance was 60% (3 out of the 5 incorrect feeders, dashed line in Fig. 2B)). Importantly, a 2 (group) $\times$ 2 (condition) ANOVA indicated no significant main effect of group ($F(1,29) = 0.327$, $p = 0.537$) or condition ($F(1,29) = 0.454$, $p = 0.507$) and no significant interaction ($F(1,29) = 0.791$, $p = 0.381$). These results suggest that aged rats had consolidated and remembered Set 1 as well as young rats at the time of the reminder on Day 2.

3.1.2. Experiment 1.1 – the role of context as a reminder

The experimental context has been shown to be a sufficient reminder to trigger reconsolidation in humans (Hupbach et al., 2008) and rodents (Artinian, De Jaeger, Fellini, de Saint Blanquat, & Roullet, 2007). In Experiment 1.1, we manipulated the context (visual, odor, and texture cues) in which the second set of feeders (Set 2) was learned in order to either remind or not remind rats of a previously learned set (Set 1). Rats were cued to recall Set 1 24 h after learning Set 2. A 2 (group) $\times$ 2 (condition) ANOVA was conducted separately on the percent correct recall and percent

Fig. 2. Learning performance. (A) Mean percent errors made during training on Days 1 and 2 in the Reminder and No Reminder conditions in the young and aged rats. (B) Mean proportion of Day 2 errors made to Set 1 feeders in the Reminder and No Reminder conditions in young and aged rats. Eight young and 15 aged rats were used to obtain these data. All error bars represent standard errors of means.

intrusions. A significant main effect of group was found for correct recall, $F(1,36) = 5.985$, $p = .019$, indicating that the young rats (Fig. 3A) had a greater proportion of visits to Set 1 feeders than the aged rats (Fig. 3B). Neither the main effect of condition ($F(1,36) = 0.494$, $p = 0.486$) nor the interaction ($F(1,36) = 0.101$, $p = 0.752$) was significant. Regarding intrusions, there was a significant main effect of group, $F(1,36) = 12.189$, $p = .001$; a significant main effect of condition, $F(1,36) = 11.148$, $p = .002$; and a significant interaction between the two, $F(1,36) = 6.701$, $p = .014$. Follow up pairwise comparisons revealed that young rats in the Reminder condition made significantly more intrusions than young rats in the No Reminder condition and significantly more than aged rats in both conditions ($p < .01$ in all cases). There was no significant difference between the percent of intrusions made by aged rats in the Reminder and No Reminder conditions, nor between young rats in the No Reminder condition and aged rats in either condition (all p's > 0.9). Analyzing errors to Set 2 feeders separately

from those to No Set feeders revealed that the difference in intrusions between conditions in young rats was driven by a difference in visits to Set 2 feeders only (Table S4). The percent of LED-cued visits (when rats did not reach the baited feeder within the time limit; not included in recall calculations) did not differ between age groups or conditions, and there was no significant interaction (2×2 ANOVA, all p's > 0.4). These results suggest that, in young rats but not in aged, re-exposure to the context prompted reconsolidation of the memory for the Set 1 learning episode, allowing for updating to occur.

3.1.3. Experiment 1.2 – the selectivity of the reminder effect

One possibility is that, rather than expressing an updated memory, the rats are simply visiting all of the feeders on which they have been trained, regardless of the set to which they belong. Experiment 1.2 assessed the specificity of the intrusion effect in aged rats. Fig. 4 shows the percent correct recall of Set 2 and intru-
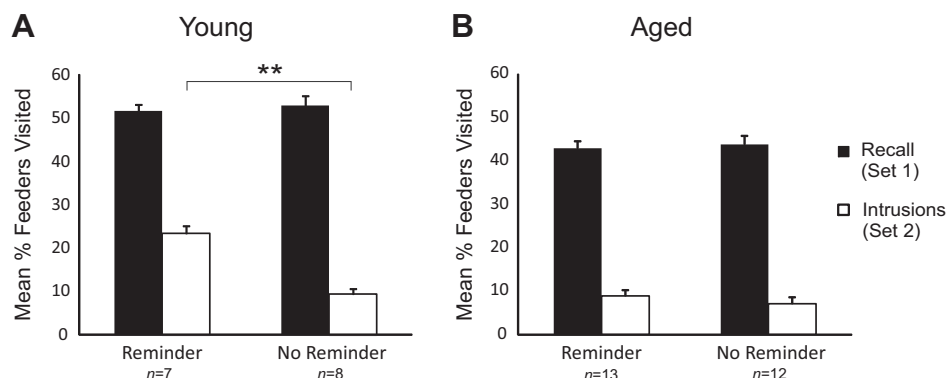
Fig. 3. Experiment 1.1. (A) Mean percentage of feeders visited belonging to Set 1 and Set 2 during recall of Set 1 on Day 3 in young rats. Eight rats were used to obtain these data. (B) Mean percentage of feeders visited belonging to Set 1 and Set 2 during recall of Set 1 on Day 3 in aged rats. Fifteen rats were used to obtain these data. All error bars represent standard errors of means.
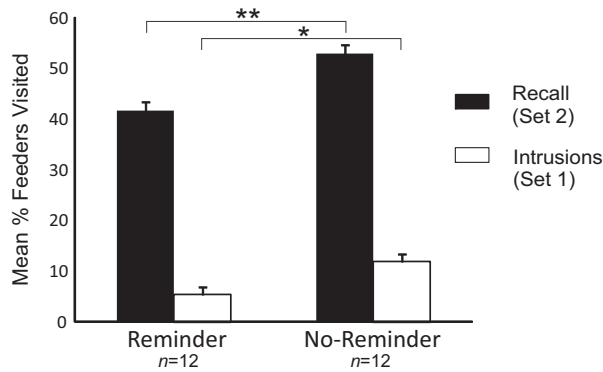
Fig. 4. Experiment 1.2. Mean percentage of feeders visited belonging to Set 1 and Set 2 during recall of Set 2 on Day 3 in aged rats. Error bars represent standard errors of means. Thirteen rats were used to obtain these data.

sions from Set 1. Unexpectedly, the No Reminder condition had both a higher percent correct recall ($t = -2.92$, $p = 0.008$) and intrusions ($t = -2.42$, $p = 0.024$) than the Reminder condition. Importantly, and in contrast to our previous results in young rats, aged rats made as many intrusions from Set 1 during Set 2 retrieval as they did from Set 2 during Set 1 retrieval (Reminder: $t = 1.166$, $p = 0.256$; No Reminder: $t = -1.559$, $p = 0.133$). These results suggest that the intrusions observed in the aged rats do not result from a reminder-triggered reconsolidation process but rather reflect general interference between the two sets.

3.1.4. Experiment 1.3 – the time course of the reminder effect

Experiment 1.3 investigated the time course of the intrusion effect in aged rats, testing recall of Set 1 either 15 min or 4 h (different groups at each time point) following Set 2 learning. Fig. 5 illustrates the percent of intrusions made at these points as well as after 24 h (data from experiment 1.1). A 3 (time) × 2 (condition) ANOVA indicated a significant main effect of time, $F(2,50) = 10.696$, $p < .001$, and follow-up pairwise comparisons revealed that intrusions declined significantly between the 15 min and 4hr points ($p = 0.005$) but not between 4 h and 24 h ($p = 0.734$). There was also a significant main effect of condition, $F(1,50) = 10.828$, $p = 0.002$, indicating more intrusions in the Reminder (black line) than the No Reminder (white line) condition. The interaction did not reach significance, $F(2,50) = 2.761$, $p = .073$. Planned pairwise comparisons revealed that rats in the Reminder condition made significantly more intrusions at the 15 min time point than rats in the No Reminder condition at this point
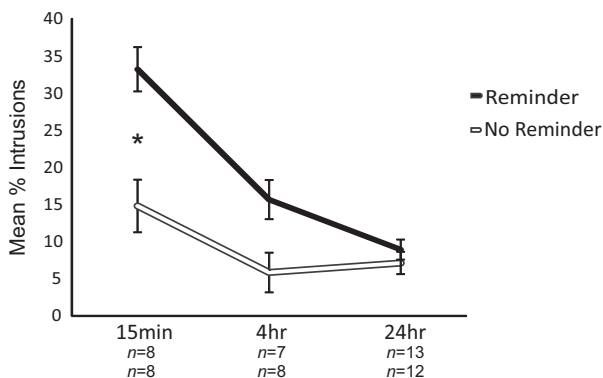
Fig. 5. Experiment 1.3. Mean percentage of Intrusions during recall of Set 1 either 15 min, 4 h, or 24 h (data from Experiment 1.1) following Set 2 training in aged rats. Fourteen rats were used to obtain the data in this experiment. All error bars represent standard errors of means.

($p = .027$). However, the difference between the conditions at 4 h was not significant ($p = 0.531$). These results indicate that the Set 2 learning context does influence memory interference in the aged rats, but only in the short-term.

3.2. Memory reconsolidation in young adult and older adult humans

In this study, we were interested in whether the age-related difference in memory performance observed in rats would extend to humans. Therefore, we tested young and older adults using the procedure of Hupbach et al. (2008), on which the rat study was based. Although episodic memory deficits have frequently been observed in normally-aging older adults, the reasons for these impairments have been ascribed not only to declines in hippocampal function but also, under some circumstances, to declines in prefrontal function. Although we expected reconsolidation effects to be associated with medial temporal function, the binding of context to content during initial encoding and at retrieval is also found to depend at least partly on prefrontal function (see source memory studies, e.g., (Glisky et al., 2001; Mitchell, Johnson, Raye, & Greene, 2004; Nolde, Johnson, & D'Esposito, 1998; Slotnick, Moo, Segal, & Hart, 2003)). We therefore decided to explore the effects of individual differences in medial temporal and prefrontal function in older adults in the reconsolidation paradigm.
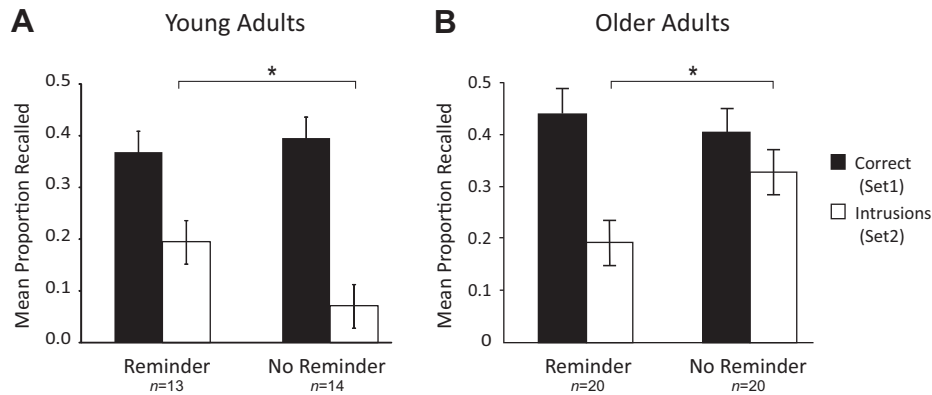
3.2.1. Young adults

We tested a control group of 27 young adults, 13 in the Reminder condition and 14 in the No-Reminder condition. Results from the young adults replicated findings from earlier studies (see Fig. 6A). On Day 3, participants in the Reminder group showed significantly more intrusions (.20) from Set 2 into recall of Set 1 than those in the No-Reminder group (.07), $t(25) = 2.12$, $p = .04$. There were no differences between groups in correct recall of Set 1 (Reminder: $M = .37$, $SD = .15$; No Reminder: $M = .40$, $SD = .19$, $t(25) < 1$). A 2 × 2 mixed ANOVA of the number of learning trials to criterion indicated only a significant main effect of Day, $F(1,23) = 11.5$, $p = .002$. People learned the new information in fewer trials on Day 2 ($M = 2.93$, $SD = 1.1$) than on Day 1 ($M = 3.63$, $SD = 1.0$). There was no significant effect of condition and no significant interaction (p's > 0.05).

3.2.2. Older adults

3.2.2.1. Learning performance on Days 1 and 2.
The number of learning trials needed for participants to recall at least 13 of the 15 items on Day 1 and Day 2 is shown in Fig. S3. Those who recalled less than 13 items on the fifth learning trial were given a score of 6. The average number of trials to reach criterion was 3.93 ($SD = 1.42$) on Day 1 and 3.75 ($SD = 1.6$) on Day 2. A 2 (day) x 2 (condition) x 2 (FL status) x 2 (MTL status) mixed ANOVA indicated only one main effect: High MTL older adults learned the lists more quickly (3.2 trials) than low MTL participants (4.5 trials), $F(1,32) = 9.66$, $p = .004$. There was also an interaction between MTL and condition, $F(1,32) = 4.38$, $p = .04$, indicating that this advantage was significant only in the Reminder condition. The only other significant effect was an interaction between day, condition, and FL status, $F(1,32) = 4.17$, $p = .049$, which reflected a tendency for individuals with high FL function to learn the material more quickly than those with low FL function only in the Reminder condition on Day 2 ($p = 0.09$).

3.2.2.2. Recall of Set 1 on Day 3.
The mean proportion of items correctly recalled from Set 1 and the mean proportion of items falsely recalled from Set 2 (i.e., intrusions) were analyzed in separate 2 (condition) × 2 (FL) × 2 (MTL) ANOVAs. For correctly recalled items, there were no significant effects (all p's > 0.05). Overall, older adults in the Reminder condition correctly recalled 44% of the

Fig. 6. Experiment 2. Mean proportion of objects correctly and falsely recalled by young adults (A) and older adults (B) on Day 3 in the aged human study. Objects that were mistakenly recalled from Set 2 are labeled as Intrusions. Error bars represent standard errors of means.

Set 1 items and those in the No Reminder condition recalled 40% (Fig. 6B, black bars). For the intrusion errors from Set 2, however, there was a significant main effect of condition, which was the opposite of that reported in previous studies with younger adults. Older adults in the No Reminder condition produced significantly more intrusions (33%) than those in the Reminder condition (19%), $F(1,32) = 4.93$, $p = .03$ (Fig. 6B, white bars). This effect did not significantly interact with group factors (p's > 0.05); the pattern was observable in all neuropsychological groups (see Fig. S4). No other effects were significant (all p's > 0.05).

4. Discussion

This study explored the effects of aging on performance in analogous context-dependent reconsolidation tasks in rats and humans. In both species, we found that younger animals in the Reminder condition made more intrusion errors than those in the No Reminder condition (Figs. 3A and 6A). That is, memory for Set 2 was more often expressed during Set 1 recall when Set 2 was learned in the same context as Set 1. This result replicates previous findings using the same tasks (Hupbach et al., 2008; Jones et al., 2012). Re-exposure to the experimental context has been shown to be sufficient to induce reactivation and reconsolidation (Artinian et al., 2007; Sara, 2010). Therefore, in young animals, re-entering the Set 1 learning context on Day 2 may have triggered reactivation of the Set 1 memory, allowing the newly learned Set 2 items to be integrated into the Set 1 memory. An alternative interpretation is that, because Set 2 was learned in the same context in which retrieval took place in the Reminder condition, more intrusions in this condition reflect stronger interference at the time of retrieval (Wixted, 2004). Both explanations rely on the context cuing a set memory, but at different times (reconsolidation on Day 2 and interference on Day 3). Though our results cannot disambiguate between these two influences, we note that context continuity between Day 1 and Day 2 had no effect on the proportion of erroneous visits made to Set 1 feeders during the test phase on Day 2. Future studies using pharmacological interventions on Day 2 could confirm that the Set 1 memory undergoes reconsolidation.

Another possibility is that intrusions resulted from Set 1 being internally reactivated during Set 2 learning, independent of the context. If so, some items of Set 1 may have been bound to Set 2 items 'directly' (i.e. not through their association to the context per se). Recent evidence however suggests that recall in and of itself may not be sufficient to trigger reconsolidation of fear memory (Sevenster, Beckers, & Kindt, 2012). Together with the similarity between conditions in the proportion of Set 1 errors during Set 2 learning, these findings suggest that this interpretation is unlike-

ly. The lack of reinforcement (rewards) of Set 1 feeders on Day 2 (i.e. a mismatch between original learning and reactivation), combined with the context, may be key in triggering labilization–reconsolidation processes (Forcato, Fernandez, & Pedreira, 2014; Nadel, 2008).

In both species, the pattern of results differed between young and aged subjects. In aged rats, there was no difference in the percent intrusions between the Reminder and No Reminder conditions (Fig. 3B). In older adult humans, there was a reversal of the young adult pattern: they produced significantly more intrusion errors in the No Reminder condition (Fig. 6B). Furthermore, there was no hint of any involvement of MTL or FL function in older adults, as measured by our factor scores, in the differential number of intrusions in the two conditions. In fact, all neuropsychological groups showed the reverse reminder effect, and there was no interaction with condition (Fig. S4).

It is important to consider whether the difference in the pattern of memory intrusions between young and aged animals in this study could have resulted from age-related impairment in learning or consolidation. We believe this explanation is unlikely for several reasons. First, regarding learning, both human and rat aged groups reached the same (rat) or an equivalent (human, 13/15 vs. 17/20 objects) learning criterion as young groups. Second, in the human experiment, despite appearing sensitive to learning abilities (as indicated by the fact that the high MTL group learned the two sets in significantly fewer trials initially than the low MTL group; Fig. S3), MTL functioning level made no difference in the pattern of intrusions (Fig. S4). In rats, when Set 1 recall took place soon after Set 2 training (Fig. 5), aged animals made many erroneous visits to Set 2 (on par with the amount seen previously in young rats (Jones et al., 2012)), indicating that they had indeed learned it. Third, regarding consolidation, aged rats made the same proportion of errors to Set 1 feeders during Set 2 training (24hrs after learning Set 1) as young rats (both groups were significantly above chance at visiting Set 1; Fig. 2B), suggesting they had consolidated Set 1 equally as well as young rats. Likewise, on Day 3, aged rats in both conditions showed the same amount of intrusions as young rats in the No Reminder condition (Fig. 3B). Older humans recalled at least as many Set 1 (correct) and Set 2 (intruding) objects on Day 3 as did young adults (Fig. 6) and (with the exception of increased intrusions in the aged No Reminder group) approximately the same amount as young adults in the original Hupbach, Gomez, Hardt, and Nadel (2007) study. Despite this evidence, we cannot completely rule out the possibility of age-related differences in learning and/or consolidation. However, we note findings by others that weaker memories are more vulnerable to reconsolidation than stronger memories (Robinson & Franklin, 2010; Suzuki, Josselyn, Frankland, Masushige, Silva, & Kida, 2004;

Wang, de Oliveira Alvares, & Nader, 2009; Winters, Tucci, & DaCosta-Furtado, 2009). Therefore, if the aged groups did not encode or consolidate the episodes to the same extent as the young groups, one might expect to see more indication of reconsolidation rather than less. Taken together, this evidence suggests that, while it cannot be completely ruled out, age-related impairment in learning and/or consolidation is unlikely to account for the findings of this study.

We next consider that reactivation and reconsolidation processes may have been active in both conditions in aged animals. Studies have shown that aging is associated with deficits in pattern separation (Yassa, Lacy, Stark, Albert, Gallagher, & Stark, 2011a; Yassa, Mattfeld, Stark, & Stark, 2011b). It is possible that the contexts used in our rat experiments were not different enough to allow the aged animals to distinguish between them. If this were the case, then it would explain why manipulating the context during Set 2 learning did not affect the performance of the rats, as well as why there was no difference between retrieval in Context A and Context C (Fig. S2). However, results of Experiment 1.3 indicate that aged rats could distinguish between the contexts. In this case, rats in the No Reminder condition made significantly fewer intrusions than in the Reminder condition 15 min after learning Set 2 (Fig. 5). These intrusions in the short-term likely reflect retrieval competition between Set 2, which had just been learned, and Set 1, which the rats were cued to recall. The context change between Set 2 learning (Context B) and Set 1 recall (Context A) for the No Reminder condition apparently reduced the retrieval competition, resulting in fewer intrusions than the Reminder condition. Therefore, the aged rats could distinguish between the different contexts, at least when they were presented close together in time. However, it is possible that they failed to notice the difference between the contexts with greater time intervals between exposures. Further studies using neurophysiological recordings in the rodent hippocampus and measuring the extent of place cell remapping in the different contexts used here could shed light on the extent to which contexts are perceived to be different on an animal to animal basis (Anderson, Killing, Morris, O'Donoghue, Onyiagha, Stevenson, Verriotis, & Jeffery, 2006). See Fig. S5 and Supplemental Information for further discussion regarding the time course of intrusions in young and aged animals.

The contexts used in the human experiment were two different rooms. However, it is possible that older adults failed to encode the two locations differentially but instead coded them non-specifically, for example as "rooms at the university." There is some evidence consistent with this possibility. Previous studies of source memory in older adults (e.g., Glisky et al., 2001) have shown that repeating the context facilitates memory for target items even though older people cannot always recall the specific context, suggesting that the context may have been encoded at a general level but without distinctive detail. If this were the case, the context may have served as a reminder in both conditions in older adults. However, the questionnaire administered at the end of the testing session indicated that all but one of the older adults were able to specify whether they were in the same or a different room on Day 1 and Day 2 and to describe some specific details of the rooms. Therefore, in both rats and humans, it seems unlikely that reconsolidation occurred in both conditions as a result of an inability to distinguish between the contexts.

Another possibility is that re-exposure to the context failed to trigger reactivation and reconsolidation in aged animals. Results of Experiment 1.2 indicate that this may indeed be the case. Regardless of whether or not the rats could distinguish the contexts in Experiment 1.1, if the intrusions were due to a specific reconsolidation effect, then we would expect more intrusions from Set 2 during Set 1 recall than from Set 1 during Set 2 recall (as Set 2 was never reactivated after it was consolidated, any intrusions

during its recall could not be the result of reconsolidation). Unlike what we observed previously in young rats (Jones et al., 2012), we found that in both the Reminder and No Reminder conditions, aged rats made as many intrusions from Set 1 into Set 2 as they did from Set 2 into Set 1 (Fig. 4). These results suggest that the intrusions made by aged animals in this study do not reflect reconsolidation but rather a general interference process. Though not tested in humans here, adding a condition in which older adults are asked to retrieve the Day 2 set rather than the Day 1 set would predict intrusions from Set 1 into Set 2 recall. However, if intrusions in older adults were attributable to a source memory problem, one might have predicted an influence of frontal function (see Glisky et al., 2001).

Why might a contextual reminder fail to trigger reconsolidation in aged animals? Studies have shown that hippocampal cells representing spatial location sometimes spontaneously remap between exposures to a familiar context in aged rats but not in young rats (Barnes, Suster, Shen, & McNaughton, 1997; Hok, Chah, Reilly, & O'Mara, 2012; Wilson, Ikonen, Gureviciene, McMahan, Gallagher, Eichenbaum, & Tanila, 2004). Furthermore, whereas young rats show remapping at the first exposure to a novel context, aged animals require more experience or multiple exposures to remap between a familiar and novel context (Wilson et al., 2004). In our study, rats in the Reminder condition learned Set 1 and Set 2 in Context A, which was a novel context compared to the familiar pre-training context. Young rats likely had a spatial representation of Context A which formed on Day 1 and remained stable across exposures, allowing for reactivation of the Set 1 memory on Day 2 and integration of Set 2 items into that representation. Aged rats, however, might have had a representation that did not fully form until Day 2 or which spontaneously remapped between the learning sessions, preventing memory reactivation and reconsolidation on Day 2. Similarly, the unique contextual details of the memory may have been lost after 24 h in aged animals, such that the context no longer acted as a strong enough cue to reactivate the representation. Another possibility is that the aged rats had a stable representation of the environment but were impaired at associating the set items (feeders) with the overall context. Such impairment could have prevented the context from being able to reactivate the set items and would explain why the retrieval context failed to influence the intrusions in the aged rats, unlike what we previously observed in young rats. It should be noted that such explanations would also pertain to age-related changes in context-dependent retrieval competition/interference. Further electrophysiological studies of the rat hippocampus in this task should be useful and could inform future fMRI studies in humans.

The basic finding in the rat experiments of an age-related difference in the pattern of memory intrusions extended to humans in this study. However, unlike in the rats, older adults in the No Reminder group made significantly more intrusion errors than those in the Reminder group (Fig. 6B). If the intrusions were due to general interference, as we suggest here, then why would the No Reminder group show more interference than the Reminder group in the human participants? One possible explanation is that learning Set 2 in a novel context (Context B rather than Context A) led the No Reminder group to consolidate Set 2 more distinctively (allowing it to be more easily recalled) than the Reminder group. If this were the case, then we would expect the No Reminder group to perform better than the Reminder group at recalling Set 2 on Day 3. Alternatively, it is possible that the presentation of the Day 1 context on Day 2 in the Reminder condition strengthened the Set 1 item-context association. On Day 3, when the context was repeated for the third time, it may have provided a stronger cue to Set 1 items than to Set 2 items, reducing the Set 2 intrusions. In the No Reminder condition, the context may not have been as

good a cue for Set 1 items, increasing the likelihood of retrieving the more recent Set 2 items on Day 3. Testing recall on the third day in a novel context might provide information relevant to this interpretation.

To our knowledge, this is the first study investigating reconsolidation in aged animals in a task closely matched to that given to humans. Spatial context plays a vital role in episodic memory processing. However, as opposed to their effect in young animals, our results suggest that contextual reminders fail to induce reactivation and reconsolidation of spatial memories in aged rats and episodic memories in aged humans. Importantly, the current study does not suggest that reconsolidation never occurs in old age, but rather that the ability of spatial contextual cues to trigger this process is diminished. Future work is needed to determine the mechanisms underlying this age difference, examine the efficacy of different types of reminder cues in aged animals, and to assess the effects of aging on reconsolidation in different memory systems. Other experimental paradigms could validate and enhance the behavioral results presented here, including the use of computational models to study the mechanisms of context-dependent reconsolidation (Lines, Nation, & Fellous, 2014), the use of protein synthesis inhibitors to block memory reconsolidation, the use of immediate early gene techniques to image the neural correlates of spatial memory items and context, and electrophysiological and optogenetic studies to analyze and manipulate hippocampal activity at different phases of the experiments.

Acknowledgments

Appendix A. Supplementary material

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.nlm.2015.02.003.

References

Anderson, M. I., Killing, S., Morris, C., O'Donoghue, A., Onyiagha, D., Stevenson, R., et al. (2006). Behavioral correlates of the distributed coding of spatial context. *Hippocampus, 16*(9), 730–742.

Artinian, J., De Jaeger, X., Fellini, L., de Saint Blanquat, P., & Roullet, P. (2007). Reactivation with a simple exposure to the experimental environment is sufficient to induce reconsolidation requiring protein synthesis in the hippocampal CA3 region in mice. *Hippocampus, 17*, 181–191.

Barnes, C. A., Suster, M. S., Shen, J., & McNaughton, B. L. (1997). Multistability of cognitive maps in the hippocampus of old rats. *Nature, 388*, 272–275.

Benton, A. L., & Hamsher, K. de S. (1976). *Multilingual aphasia examination manual.* Iowa City: University of Iowa.

Bower, M. R., Euston, D. R., & McNaughton, B. L. (2005). Sequential-context-dependent hippocampal activity is not necessary to learn sequences with repeated elements. *Journal of Neuroscience, 25*, 1313–1323.

Burke, S. N., & Barnes, C. A. (2006). Neural plasticity in the aging brain. *Nature Reviews Neuroscience, 7*, 30–40.

Delis, D. C., Kramer, J., Kaplan, E., & Ober, B. A. (1987). *The California verbal learning test.* San Antonio: The Psychological Corporation.

Folstein, M. F., Folstein, S. F., & McHugh, P. R. (1975). Mini-mental state: A practical method for grading the cognitive state of patients for the clinician. *Journal of Psychiatric Research, 12*, 189–198.

Forcato, C., Fernandez, R. S., & Pedreira, M. E. (2014). Strengthening a consolidated memory: The key role of the reconsolidation process. *Journal of Physiology-Paris*

(14). http://dx.doi.org/10.1016/j.jphysparis.2014.09.001, pii: S0928-4257(14)00044-8.

Glisky, E. L., & Kong, L. L. (2008). Do young and older adults rely on different processes in source memory tasks? A neuropsychological study. *Journal of Experimental Psychology. Learning, Memory, and Cognition, 34*, 809–822.

Glisky, E. L., Rubin, S. R., & Davidson, P. S. (2001). Source memory in older adults: An encoding or retrieval problem? *Journal of Experimental Psychology. Learning, Memory, and Cognition, 27*, 1131–1146.

Hart, R. P., Kwentus, J. A., Wade, J. B., & Taylor (1988). Modified Wisconsin Card Sorting Test in elderly normal, depressed, and demented patients. *Clinical Neuropsychologist, 2*, 49–56.

Hok, V., Chah, E., Reilly, R. B., & O'Mara, S. M. (2012). Hippocampal dynamics predict interindividual cognitive differences in rats. *Journal of Neuroscience, 32*, 3540–3551.

Hupbach, A., Gomez, R., Hardt, O., & Nadel, L. (2007). Reconsolidation of episodic memories: A subtle reminder triggers integration of new information. *Learning & Memory, 14*, 47–53.

Hupbach, A., Gomez, R., & Nadel, L. (2009). Episodic memory reconsolidation: Updating or source confusion? *Memory, 17*(5), 502–510.

Hupbach, A., Hardt, O., Gomez, R., & Nadel, L. (2008). The dynamics of memory: Context-dependent updating. *Learning & Memory, 15*, 574–579.

Jones, B., Bukoski, E., Nadel, L., & Fellous, J. M. (2012). Remaking memories: Reconsolidation updates positively motivated spatial memory in rats. *Learning & Memory, 19*, 91–98.

Lines, J., Nation, K., & Fellous, J. M. (2014). A connectionist model of context-based memory reconsolidation in the hippocampus: The role of sleep. *BMC Neuroscience, 15*(Suppl 1, CNS 2014), 163.

Mitchell, K. J., Johnson, M. K., Raye, C. L., & Greene, E. J. (2004). Prefrontal cortex activity associated with source monitoring in a working memory task. *Journal of Cognitive Neuroscience, 16*, 921–934.

Nadel, L. (2008). The hippocampus and context revisited. In S. J. Y. Mizumori (Ed.), *Hippocampal place fields: Relevance to learning and memory* (pp. 3–15). New York: Oxford University Press.

Nader, K., & Einarsson, E. O. (2010). Memory reconsolidation: An update. *Annals of the New York Academy of Sciences, 1191*, 27–41.

Nolde, S. F., Johnson, M. K., & D'Esposito, M. (1998). Left prefrontal activation during episodic remembering: An event-related fMRI study. *NeuroReport, 9*, 3509–3514.

Rajah, M. N., Kromas, M., Han, J. E., & Pruessner, J. C. (2010). Group differences in anterior hippocampal volume and in the retrieval of spatial and temporal context memory in healthy young versus older adults. *Neuropsychologia, 48*(14), 4020–4030.

Robinson, M. J., & Franklin, K. B. (2010). Reconsolidation of a morphine place preference. impact of the strength and age of memory on disruption by propranolol and midazolam. *Behavioural Brain Research, 213*, 201–207.

Sara, S. J. (2000). Retrieval and reconsolidation: toward a neurobiology of remembering. *Learning & Memory, 7*, 73–84.

Sara, S. J. (2010). Reactivation, retrieval, replay and reconsolidation in and out of sleep: Connecting the dots. *Frontiers in Behavioral Neuroscience, 4*, 185.

Sevenster, D., Beckers, T., & Kindt, M. (2012). Retrieval per se is not sufficient to trigger reconsolidation of human fear memory. *Neurobiology of Learning and Memory, 97*(3), 338–345.

Slotnick, S. D., Moo, L. R., Segal, J. B., & Hart, J. Jr., (2003). Distinct prefrontal cortex activity associated with item memory and source memory for visual shapes. *Brain Research Cognitive Brain Research, 17*, 75–82.

Suzuki, A., Josselyn, S. A., Frankland, P. W., Masushige, S., Silva, A. J., & Kida, S. (2004). Memory reconsolidation and extinction have distinct temporal and biochemical signatures. *Journal of Neuroscience, 24*, 4787–4795.

Tronson, N. C., & Taylor, J. R. (2007). Molecular mechanisms of memory reconsolidation. *Nature Reviews Neuroscience, 8*, 262–275.

Wang, S. H., de Oliveira Alvares, L., & Nader, K. (2009). Cellular and systems mechanisms of memory strength as a constraint on auditory fear reconsolidation. *Nature Neuroscience, 12*, 905–912.

Wechsler, D. (1981). *Wechsler adult intelligence scale – Revised.* New York: Psychological Corporation.

Wechsler, D. (1987). *Wechsler memory scale – Revised.* New York: Psychological Corporation.

Wechsler, D. (1997). *Wechsler memory scale – III manual.* San Antonio: Psychological Corporation.

Wilson, I. A., Ikonen, S., Gureviciene, I., McMahan, R. W., Gallagher, M., Eichenbaum, H., et al. (2004). Cognitive aging and the hippocampus: How old rats represent new environments. *Journal of Neuroscience, 24*, 3870–3878.

Winters, B. D., Tucci, M. C., & DaCosta-Furtado, M. (2009). Older and stronger object memories are selectively destabilized by reactivation in the presence of new information. *Learning & Memory, 16*, 545–553.

Wixted, J. T. (2004). The psychology and neuroscience of forgetting. *Annual Review of Psychology, 55*, 235–269.

Yassa, M. A., Lacy, J. W., Stark, S. M., Albert, M. S., Gallagher, M., & Stark, C. E. (2011a). Pattern separation deficits associated with increased hippocampal CA3 and dentate gyrus activity in nondemented older adults. *Hippocampus, 21*, 968–979.

Yassa, M. A., Mattfeld, A. T., Stark, S. M., & Stark, C. E. (2011b). Age-related memory deficits linked to circuit-specific disruptions in the hippocampus. *Proceedings of the National Academy of Sciences of the United States of America, 108*, 8873–8878.