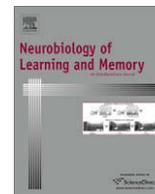


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Reconsolidation in humans opens up declarative memory to the entrance of new information

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ABSTRACT

A consolidated memory recalled by a reminder enters a vulnerability phase (labilization), followed by a process of stabilization (reconsolidation). Several authors have suggested that the labilization of the consolidated memory makes the incorporation of new information possible. Here, we demonstrate updating in the framework of memory declarative reconsolidation in humans by giving an opportune verbal instruction. Volunteers learn an association between five cue-syllables (L1) and their respective response-syllables. Twenty-four hours later, the paired-associate verbal memory is labilized by exposing the subjects to the reminder, and then they receive the verbal instruction of adding three new cue-response syllables (INFO) with their respective responses to the former list of five. The new information is incorporated into the single former L1-memory and both INFO and L1 are successfully retrieved on the third day. However, when the instruction is not preceded by a proper reminder, or when the instruction omits the order of adding the INFO into the former L1-memory, we observed interference in retrieval of both the original and the new information, suggesting that they are encoded independently and coexist as separate memories.

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1. Introduction

According to the reconsolidation hypothesis, consolidated memory recalled by a reminder enters a vulnerability phase (labilization) during which it is transiently sensitive to disruption (or enhancement), followed by a process of stabilization that returns memory to the former state (Nader, Schafe, & LeDoux, 2000; Sara, 2000a). The reminder is the event that triggers the labilization–reconsolidation process of the memory. This process has been shown in very diverse species and types of memory, including the human procedural memory of a motor skill task (Walker, Brakefield, Hobson, & Stickgold, 2003), and reconsolidation in verbal learning (Forcato, Burgos, Argibay, Pedreira, and Maldonado, 2007). In both cases, the amnesic agent was other learning and recently, Kindt, Soeter, and Vervliet (2009) showed that oral administration of the β -adrenergic receptor antagonist propranolol disrupts the human labilization–reconsolidation process of a fear memory, erasing its behavioural expression 24 h later and preventing the return of fear.

Since the early formulation of the reconsolidation hypothesis, several controversial items have arisen, mainly concerning the

dubious functionality of positing a consolidated memory in the transient but risky stage of being labilized. Several authors proposed that retrieval triggers a reconsolidation process that allows the integration of new information into the background of the past (memory updating). In other words, reconsolidation would make it possible to associate new learning with already established and reactivated memories (Lewis, 1979; Nader et al., 2000; Sara, 2000b). Indeed, it seems intuitively obvious that memory needs to be continuously updated with new learning (Alberini, 2007) and some authors have studied memory updating in the framework of reconsolidation (Hupbach, Gomez, Hardt, & Nadel, 2007; Morris et al., 2006; Rodriguez-Ortiz, De la Cruz, Gutierrez, & Bermudez-Rattoni, 2005; Tronel, Milekic, & Alberini, 2005).

In our first paper on reconsolidation with humans (Forcato et al., 2007) we highlighted the suitability of this model to study the role of reconsolidation in memory updating thanks to the option of using a verbal instruction as a tool in the experiment. In the present paper, we address the possibility that a proper verbal instruction, given contingently upon a memory that has just been labilized by a reminder, allows us to add new information to the former memory.

Based on the results of our previous papers (Forcato, Argibay, Pedreira, & Maldonado, 2009; Forcato et al., 2007), the present research was aimed at studying the effect of changing either the

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parametrical conditions of the reminder or the type of verbal instruction on the updating process, in the framework of human reconsolidation.

Here, we perform a series of experiments in which volunteers learn on the first day an association between five cue-syllables and their respective response-syllables (List 1, L1). On the second day, the paired-associate verbal memory is labilized by exposing the subjects to the reminder, and then they receive the verbal Instruction of adding three new pairs of cue-response syllables (INFO) to the former list of five. The new information is incorporated into the single former L1-memory and both INFO and L1 are successfully retrieved on the third day. However, when the Instruction is not preceded by a proper reminder, or the instruction omits the order of adding the INFO to the former L1-memory, the new information is encoded in a new INFO-memory that coexists with the previous L1-memory. This co-existence produces the simultaneous recruitment of related information at retrieval, and consequently the evocation of either of the two memories interferes with the expression of the other (retrieval interference, Forcato et al., 2007, 2009; McGeoch, 1932; Greene, 1992) which brings about a poor recall of both INFO and L1. Therefore, we show the addition of new information into the former memory occurs one day after acquisition provided the information is given immediately after the reminder that triggers reconsolidation and the Instruction includes the order of adding the INFO-pairs in the L1-memory.

2. Materials and methods

2.1. Subjects

109 healthy undergraduate and graduate students from Buenos Aires University volunteered for the study. Of these, 21 participants were excluded from the data analysis because they did not reach the inclusion criteria. Their ages ranged from 20 to 35, with a mean of 25 (31 men, 57 women). Each participant was randomly assigned to one of eight groups. All participants provided written informed consent that had been approved by the Comité de Ética de la Sociedad Argentina de Investigación Clínica Review Board.

2.2. Inclusion criteria

Subjects with at least 65% of correct responses in the last four training trials (13/20 correct responses) were included in the data analysis.

2.3. Experimental room and experimental protocol

Experiments took place in a dark room and were conducted using a personal computer. Each subject was provided with ear-

phones and seated facing a monitor placed in front of a large screen on the back wall (Forcato et al., 2007, 2009).

Each three-day experiment consisted of a training session (Day 1), a treatment session (Day 2) and a testing session (Day 3). Eight distinct groups of volunteers were formed: four main groups and four control groups. The four main groups were the Reminder-Group, the No-reminder-Group, the Fake-reminder-Group and the No-instruction-Group. The four control groups were the Reminder-Control, the No-reminder-Control, the Fake-reminder-Control and the No-instruction-Control (Table 1).

2.4. The four main groups

2.4.1. The Reminder-group ($n = 12$)

2.4.1.1. The training session (Day 1). Each training trial (Fig. 1A.1) was comprised of a first period during which the context was formed (**the context period**), followed by a second during which a series of nonsense-syllables were presented as paired-associates (**the syllable period**). The context period consisted of a fixed sequence of three accumulative steps: a first step of a red light projected on the large screen for 2 s, a second step of the same light plus an image of a forest in autumn on the monitor screen for 2 s, and a third step of the light plus the image plus classical music coming through the earphones for 4-s. This specific context persisted during the syllable presentation. The syllable period that followed the former started with the presentation of a cue-syllable on the left-hand side of the monitor screen and an empty response-box on the right-hand side. Each cue-syllable was taken at random from a list of five pairs. Subjects were given 5 s to write the corresponding response-syllable. Once that period had finished, three situations were possible: first, if no syllable was written, the correct one was shown for 4 s; second, if an incorrect syllable was written, it was replaced by the correct one and it was shown for 4 s; and third, if the correct response was given, it stayed for 4 s longer. Immediately after that, another cue-syllable was shown and the process was repeated until the list was over. Altogether a trial lasted 53 s (8 s for context period and 45 s for syllable presentation). Throughout this experiment, every time a subject faced a cue-syllable and wrote down an erroneous response or no response an error was computed. The **training** consisted of the presentation of 10 trials, separated by a 4-s intertrial interval. In the first training trial, the list 1 (L1) was shown, and in the successive trials subjects were required to write down the corresponding response-syllable for each cue-syllable presented. L1 was composed of five pairs of nonsense cue-response-syllables in Spanish: **ITE-OBN**, **ASP-UOD**, **FLI-AIO**, **NEB-FOT**, **COS-GLE** (bold type: cue-syllable; regular type: response-syllable) (Fig. 1A.2). Subjects that failed to obtain 65% (13/20) of correct responses in the last four trials were excluded (Forcato et al., 2007, 2009). The training session lasted 10 min.

Table 1
Experimental Groups. L1-training stands for the L1-training received on Day 1, Instruction for the instruction to incorporate the new syllable-pairs into the L1-memory, No-instruction stands for an instruction without the order of incorporating INFO in the L1-memory, R for the reminder, R_{fake} for the fake-reminder, INFO for the info trial (specific context plus three new pair of syllables), INFO + L1-testing for the evaluation of the eight pairs of syllables (5 L1 + 3 INFO) presented at random order and L1-testing for the evaluation of the 5 L1-pairs.

	Groups	Day 1	Day 2	Day 3
Main groups	Reminder-Group	L1-training	R__Instruction__INFO	L1 + INFO-testing
	No-reminder-Group	L1-training	Instruction__INFO	L1 + INFO-testing
	Fake-reminder-Group	L1-training	R _{fake} __Instruction__INFO	L1 + INFO-testing
	No-instruction-Group	L1-training	R__No-instruction__INFO	L1 + INFO-testing
Control groups	Reminder-Control	L1-training	R	L1-testing
	No-reminder-Control	L1-training	Instruction__INFO	L1-testing
	Fake-reminder-Control	L1-training	R _{fake} __Instruction__INFO	L1-testing
	No-instruction-Control	L1-training	R__No-instruction__INFO	L1-testing

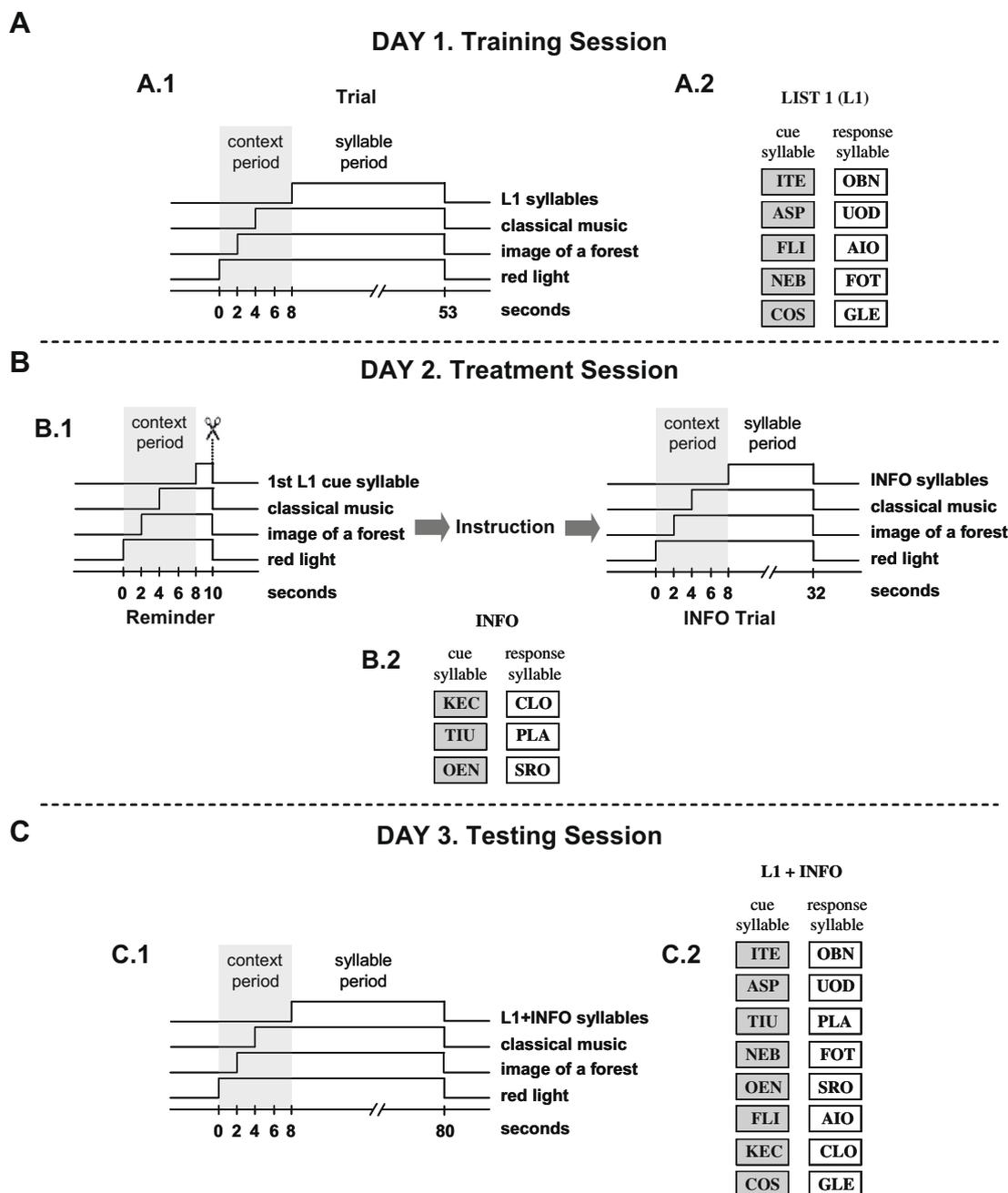


Fig. 1. Experimental protocol. (A) Day 1, training session. (A.1) A training trial was formed by the context period: a sequential and accumulative presentation of stimuli (color illumination of the room, a picture on the monitor and melody from earphones); and by the syllable period: five pairs of cue-response-syllables were presented in random order. The training session consisted of 10 trials presentation. (A.2) The list presented in the L1-training. (B) Day 2, treatment session. (B.1) The reminder includes the L1-context followed by the presentation of one L1-cue-syllable after which the trial was interrupted. After that, subjects received a specific instruction in order to incorporate the new information and the INFO trial was given. It consisted of the presentation of the L1-context followed by three new pairs of syllables. Scissor stands for the fullstop of the reminder. (B.2) The list presented in the INFO trial. (C) Day 3, testing session (C.1) The testing session consisted of two trials presentation. Each trial was formed by the context period followed by the presentation of the L1-List mixed with the INFO-list, each cue-syllable presented in a random order. (C.2) The List of eight pairs of syllables tested on Day 3.

2.4.1.2. The treatment session (Day 2). The session started with the **reminder**. The reminder trial began with the training context and immediately after its presentation, as expected, a cue-syllable appeared on the left-hand side of the monitor screen and the response-box on the right. However, 2 s later a notice displayed on the monitor announced that the session had to be suspended, thus not giving the subject time to write down the response-syllable (Fig. 1B.1). Up to this point, the experimental procedure used in this paper was substantially like those of Forcato et al. (2007, 2009), which showed in all the cases a transient labilization of the L1-mem-

ory for nearly 6 h after the reminder. In the present experiment, after the reminder presentation, the subject was instructed by the experimenter to incorporate three new pairs of syllables. The experimenter explained the **Instruction** as follows: “**Now, you are going to incorporate three new pairs of syllables to the task that you learned yesterday. These pairs of syllables are going to be presented only once. You just have to read the three pairs, you don’t have to write down any response-syllable.**” Finally, the INFO trial followed immediately after the Instruction (Fig. 1B.1). It consisted of one presentation in which subjects were only allowed to read

the syllable-pairs. It included the training context followed by a cue-syllable that appeared for 5 s, after which the corresponding response-syllable was presented and subjects were given 3 s to read the pair. Immediately after that, another cue-syllable was shown and the process was repeated until the list was over. Each cue-syllable was taken at random from a list of three pairs (Fig. 1B.2). The INFO List was composed of three pairs of nonsense cue-response-syllables: **OEN-SRO**, **TIU-PLA**, **KEC-CLO** (bold type: cue-syllable; normal type, response-syllable). The INFO trial lasted 32 s.

2.4.1.3. The testing session (Day 3). The session consisted of two test-trials separated by 4-s intertrial-interval (Fig. 1C.1). In each trial the List of eight pairs of syllables was evaluated (five pairs corresponding to L1 and three pairs corresponding to INFO) in a random order (Fig. 1C.2). Two test trials were performed to evaluate the retention in the first test trial and the effect of the retraining in the second one. The testing session lasted 3 min.

2.4.2. The No-reminder-group (n = 12)

2.4.2.1. The training session (Day 1). It was the same as the testing session of the above Reminder-Group and like that of all the remaining groups of this paper. Namely, volunteers learned an association between five **cue-syllables** and their respective **response-syllables** (List 1, L1) (Fig. 1A).

2.4.2.2. The treatment session (Day 2). It did not include the reminder but started straightforwardly with the **Instruction** followed by the INFO trial (Fig. 1B).

2.4.2.3. The testing session (Day 3). It was the same as the testing session of the Reminder-Group (Fig. 1C).

2.4.3. The Fake-reminder-group (n = 12)

2.4.3.1. The training session (Day 1). It was the same as the training session of the Reminder-Group (Fig. 1A).

2.4.3.2. The treatment session (Day 2). It was like that of the Reminder-Group but with the termed **fake-reminder** instead of the reminder. The **fake-reminder** trial began with the training context and immediately after its presentation, as expected, a cue-syllable appeared on the left-hand side of the monitor screen but in this case and unlike the reminder, **subjects were allowed to answer with the respective response-syllable** in the response-box on the right-hand side of the monitor. After that, the notice on the monitor announcing “the session has to be suspended” is displayed and it continues as in the Reminder-Group with the **Instruction** and the **INFO-trial**. (The fake-reminder was previously used in Forcato et al. (2009) and showed no labilization of the memory due to the presence of the reinforcement, i.e. absence of mismatch).

2.4.3.3. The testing session (Day 3). It was the same as the testing session of the Reminder-Group (Fig. 1C).

2.4.4. The No-instruction-group (n = 12)

2.4.4.1. Training session (Day 1). Like that of the Reminder-Group.

2.4.4.2. The treatment session (Day 2). It was like that of the Reminder-Group in all respects but omitting the usual **Instruction** and replacing it with one that omits the order of adding the INFO to the former L1: **“Now, you are going to read three pairs of syllables. These pairs of syllables are going to be presented only once. You just have to read the three pairs, you don’t have to write down any response-syllable.”**

2.4.4.3. The testing session (Day 3). Like that of the Reminder-Group (Fig. 1C).

2.5. The four control groups

2.5.1. The reminder-Control (n = 10)

2.5.1.1. The training session (Day 1). It was like that of the main groups.

2.5.1.2. The treatment session (Day 2). It consisted of the **reminder** only, namely, the reminder was not followed by the Instruction nor the INFO-trial.

2.5.1.3. The testing session (Day 3). The session consisted of two test-trials separated by 4-s intertrial-interval. In each trial the tested List consisted of the five pairs of syllables corresponding to L1 (the three pairs corresponding to INFO were not tested) presented in a random order. The testing session lasted 1.9 min.

The purpose of this group is to control the possible amnesic effect that the segment Instruction + INFO on Day 2 could have in the Reminder-Group on the L1-memory.

2.5.2. The No-reminder-Control (n = 10)

2.5.2.1. The training session (Day 1). It was like that of the main groups.

2.5.2.2. The treatment session (Day 2). It consisted of the **Instruction** and the **INFO-trial** only, namely, the reminder was not included.

2.5.2.3. The testing session (Day 3). Like that of the Reminder-Control.

The purpose of this group is to control the possible enhancing effect that testing L1- and INFO-pairs in a same session of the No-reminder-Group could have on the number of errors committed while responding to the respective cue-syllables.

2.5.3. The Fake-reminder-Control (n = 10)

2.5.3.1. The training session (Day 1). It was like that of the main groups.

2.5.3.2. The treatment session (Day 2). It consisted of the Fake-reminder presentation followed by the instruction and the INFO trial.

2.5.3.3. The testing session (Day 3). Like that of the Reminder-Control.

The purpose of this group is to control the possible enhancing effect that testing L1- and INFO-pairs in a same session of the Fake-reminder-Group could have on the number of errors committed while responding to the respective cue-syllables.

2.5.4. The No-instruction-Control (n = 10)

2.5.4.1. The training session (Day 1). It was like that of the main groups.

2.5.4.2. The treatment session (Day 2). It consisted of the reminder followed by the the **Instruction** that omits the order of adding the INFO to the former L1.

2.5.4.3. The testing session (Day 3). Like that of the Reminder-Control.

The purpose of this group is to control the possible enhancing effect that testing L1- and INFO-pairs in a same session of the No-instruction-Group could have on the number of errors committed while responding to the respective cue-syllables.

2.6. Demo

Before the training session of any group, participants were confronted with a demo program to receive all the instructions and to understand the objective of the task. The program consisted of four trials, similar in structure to those of L1 or INFO, but with another context and two different pairs of nonsense-syllables.

2.7. Statistics

2.7.1. Training performance

The uniformity of training performance was analysed with ANOVA repeated measures. Mean total number of errors in the four last training-trials was analysed with one Way-ANOVA.

2.7.2. Testing performance

Results were reported as percentage number of errors per trial for the L1 or INFO. Data from each experiment was first analyzed with a one-way analysis of variance (ANOVA). It was followed by a *a priori* planned comparisons (FISHER, $\alpha = 0.05$) between main groups.

3. Results

In order to evaluate the possibility of memory updating a three-day experiment was done. On Day 1, subjects learned a list of syllable-pairs (L1). On Day 2, they received a treatment session (presence or absence of the Reminder, presence or absence of the Instruction and presence or absence of the new list, INFO) and were tested on Day 3. Four main groups and their respective control groups were done (see Section 2).

3.1. Uniformity of training performance

A remarkable uniformity between the main groups at training was revealed by an ANOVA of repeated measures (Fig. 2, $F_{3,44} = 0.735$, $P = 0.537$) as well as no group \times trial interaction ($F_{24,352} = 0.922$, $P = 0.572$). Moreover, the analysis of the percentage of correct responses for the last four training trials disclosed no

significant difference among the groups at the training session, ($F_{3,44} = 0.433$, $P = 0.730$) (Fig. 2 inset).

3.2. Main groups performance on Day 3

The performance on Day 3 of each group was estimated by the percentage of errors committed when responding to the cue-syllables for each of the two trials separately. Subjects that received the reminder followed by the Instruction presented a better performance than those that were previously given no reminder or a fake-reminder, or than those that received an instruction which omits the order of adding the INFO to the former L1 (Fig. 3A). Specifically, the Reminder-Group made fewer percentage errors in the **new cue-syllables** (INFO) than the other three groups at the first test trial (Fig. 3A.1, $F_{3,44} = 4.377$, $P = 0.009$; Fisher-LSD $P = 0.005$, $P = 0.012$, $P = 0.002$, respectively) but no significant differences at the second test trial were disclosed (Fig. 3A.2, $F_{3,44} = 2.025$, $P = 0.124$). It is worthwhile noting that the No-reminder-Group, the Fake-reminder-Group and the No-instruction-Group failed on 89, 86 and 92% of the occasions, respectively, at test trial 1 (Fig. 3A.1). Regarding the five **former cue-syllables** (L1), the Reminder-Group made fewer percentage errors than the other two groups at the second test trial (Fig. 3B.2, $F_{3,44} = 3.644$, $P = 0.020$; Fisher-LSD $P = 0.005$, $P = 0.014$, $P = 0.014$, respectively) but a similar number at the first test trial (Fig. 3B.1, $F_{3,44} = 1.401$, $P = 0.254$).

3.3. The inclusion of the Instruction + INFO segment after the reminder has no amnesic effect on the L1-memory

The low number of errors of the Reminder-Group was similar to that of the Reminder-Control, that is, similar to a group that had the reminder but **not** the Instruction + INFO segment after reminder (Fig. 4A.1 and A.2, percentage of errors, first trial, $F_{1,20} = 0.108$, $P = 0.745$ and second trial, $F_{1,20} = 0.477$, $P = 0.498$). Therefore, the inclusion of the Instruction + INFO segment seems not to have an amnesic effect on the retention of the L1-memory, unlike previous experiments where the presentation of new training after reminder blocked the L1-memory (Forcato et al., 2007, 2009).

There was uniformity of training performance between the Reminder-Group and the Reminder-Control-Group (percentage of

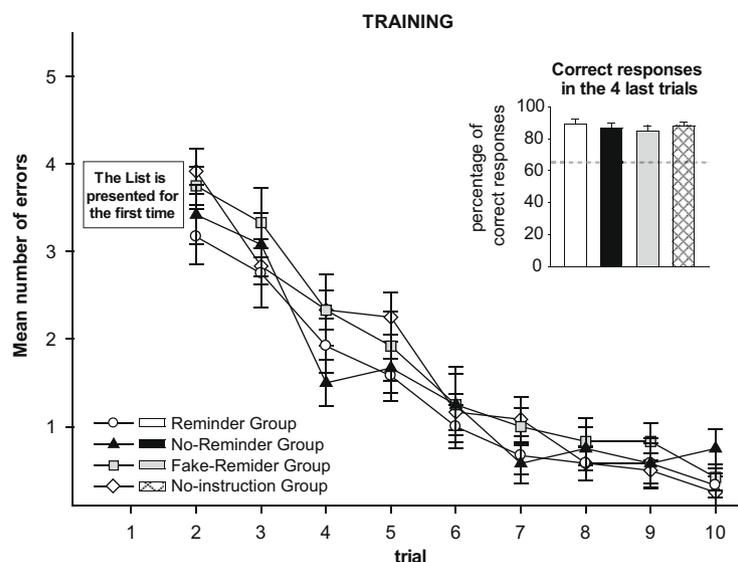


Fig. 2. L1-List training. Groups ($n = 12$). White circles stand for Reminder-Group, black triangles for No-reminder-Group, grey squares for Fake-reminder-Group, and white rhombus for No-instruction-Group. In the first trial, subjects read the list for the first time. Data presented as mean number of errors \pm SEM per trial on Day 1. Inset. White bar stands for the Reminder-Group, black bar for the No-reminder-Group, grey bar for Fake-reminder-Group and cross-hatched bar for No-instruction-Group. Mean percentage of correct responses in the four last training trials. 65% of correct responses, inclusion criteria.

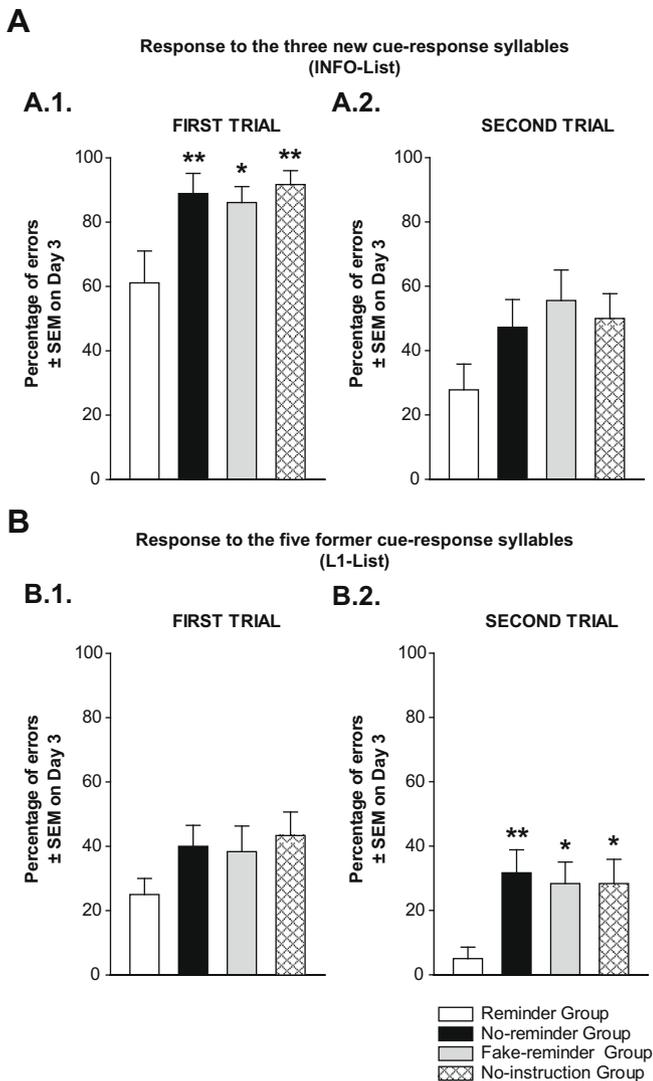


Fig. 3. Incorporation of new information. Groups ($n = 12$). (A) *INFO-list analysis*. White bars stand for the Reminder-Group, black bars for the No-reminder-Group, grey bars for Reminder-fake-Group, cross-hatched bar for No-instruction-Group. ** $p < 0.01$; * $p < 0.05$. (A.1) First test-trial. (A.2) Second test-trial. (B) *L1-list analysis*. Symbols as above. (B.1) First test-trial. (B.2) Second test-trial. All data presented as Percentage of errors \pm SEM on Day 3.

correct responses in the four last training trials, 89 ± 3 and 88 ± 3 , respectively, $F_{1,20} = 0.074$, $P = 0.788$).

3.4. Retrieval interference: the co-existence of L1 and INFO as two separate memories

The high number of errors of the No-reminder Group, the Fake-reminder-Group and the No-instruction-Group were similar to those of their respective control-Groups, that is, similar to groups where the L1-pairs but not the INFO-pairs were tested on Day 3 (Fig. 4B.1 and B.2 percentage of errors, first trial, $F_{1,20} = 1.029$, $P = 0.322$ and second trial, $F_{1,20} = 0.654$, $P = 0.428$; Fig. 4C.1 and C.2 percentage of errors, first trial, $F_{1,20} = 0.265$, $P = 0.613$ and second trial, $F_{1,20} = 0.613$, $P = 0.443$; Fig. 4D.1 and D.2; percentage of errors, first trial, $F_{1,20} = 0.080$, $P = 0.780$ and second trial, $F_{1,20} = 0.305$, $P = 0.587$). Therefore, the fact that neither the No-reminder-Group nor the Fake-reminder-Group nor the No-instruction-Group showed a higher number of L1 errors relative to those of the Reminder-Group, seems difficultly attributable to

the circumstance that L1-pairs and INFO-pairs were jointly tested on Day 3. Instead, these failures in retrieving the L1-pairs, as well as the INFO-pairs, may be explained by the fact that for the main groups, apart from the Reminder-Group, the INFO on Day 2 was encoded in a new INFO-memory that coexists with the previous L1-memory and, consequently, the evocation of either of the two memories can recruit items from the other and produce retrieval interference (McGeoch, 1932; Forcato et al., 2007, 2009; Van Dyke & McElree, 2006). On the contrary, the good performance of the Reminder-Group, concerning the INFO and L1-pairs as well as the absence of retrieval interference on the retrieval of both new and former pairs, indicates that in this group the new information on Day 2 is actually incorporated in only one memory, namely, the early L1-memory.

There was uniformity of training performance between the No-reminder-Group and the No-reminder-Control (percentage of correct responses in the four last training trials, 87 ± 3 and 85 ± 4 , respectively, $F_{1,20} = 0.128$, $P = 0.724$), between the Fake-reminder-Group and its Control (85 ± 3 and 85 ± 4 , respectively, $F_{1,20} = 0.007$, $P = 0.936$) and between the No-instruction-Group and its Control (88 ± 3 and 86 ± 4 , respectively, $F_{1,20} = 0.194$, $P = 0.664$).

4. Discussion

The main conclusion from this paper could be summed up in the following points. Firstly, when memory is labilized by the presentation of the reminder and, immediately after that, the verbal instruction and the new information are given, the subjects could introduce this new information into the recalled human memory (memory updating). Secondly, in the absence of the reminder, the new information is not incorporated into the single L1-memory although the verbal instruction is given, but it is encoded in a new INFO-memory that coexists with the former and, consequently, both memories interfere with each other at retrieval. Thirdly, the use of a fake-reminder, namely, a “reminder” that includes one syllable-response in its structure, hinders the addition of INFO into the L1-memory, confirming that the updating only occurs when the parametrical conditions of reconsolidation are fulfilled (Forcato et al., 2009). Fourthly, the omission of the explicit order of adding INFO in the instruction hinders the L1-memory updating.

It is worthwhile highlighting that, intriguingly, only a 32-s trial of the three new cue-response syllables (INFO; Fig. 1B) is enough in the Reminder-Group for the incorporation of the new information into the early L1-memory as well as for obtaining a good recall at testing 24 h later. In contrast, results of previous experiments (Forcato et al., 2009) report that ten training trials are necessary, which altogether entails a total of 15 min to obtain a performance similar to that of the Reminder-Group at testing 24 h afterwards. That is, the labilization of the L1-memory by the precedent reminder not only makes it possible to incorporate new information into the early L1-memory, but also to do it in much less time than in current training.

The finding that the Fake-reminder-Group fails to incorporate INFO into the L1-memory merits a further comment. Recently (Forcato et al., 2009) we proposed that reconsolidation is characterized by two diagnostic features: the labilization of the reactivated memory and the specificity of the reminder structure. Previous results in experiments with crabs and humans – and partially with rats – agree on pointing out that the absence of reinforcement (the mismatching component, Morris et al., 2006) is a crucial condition to define the specific structure of the reminder. Thus, the injection of high doses of cycloheximide in crabs, given after reactivation, has an amnesic effect on the context-signal-memory provided the reinforcement is not presented at any moment during

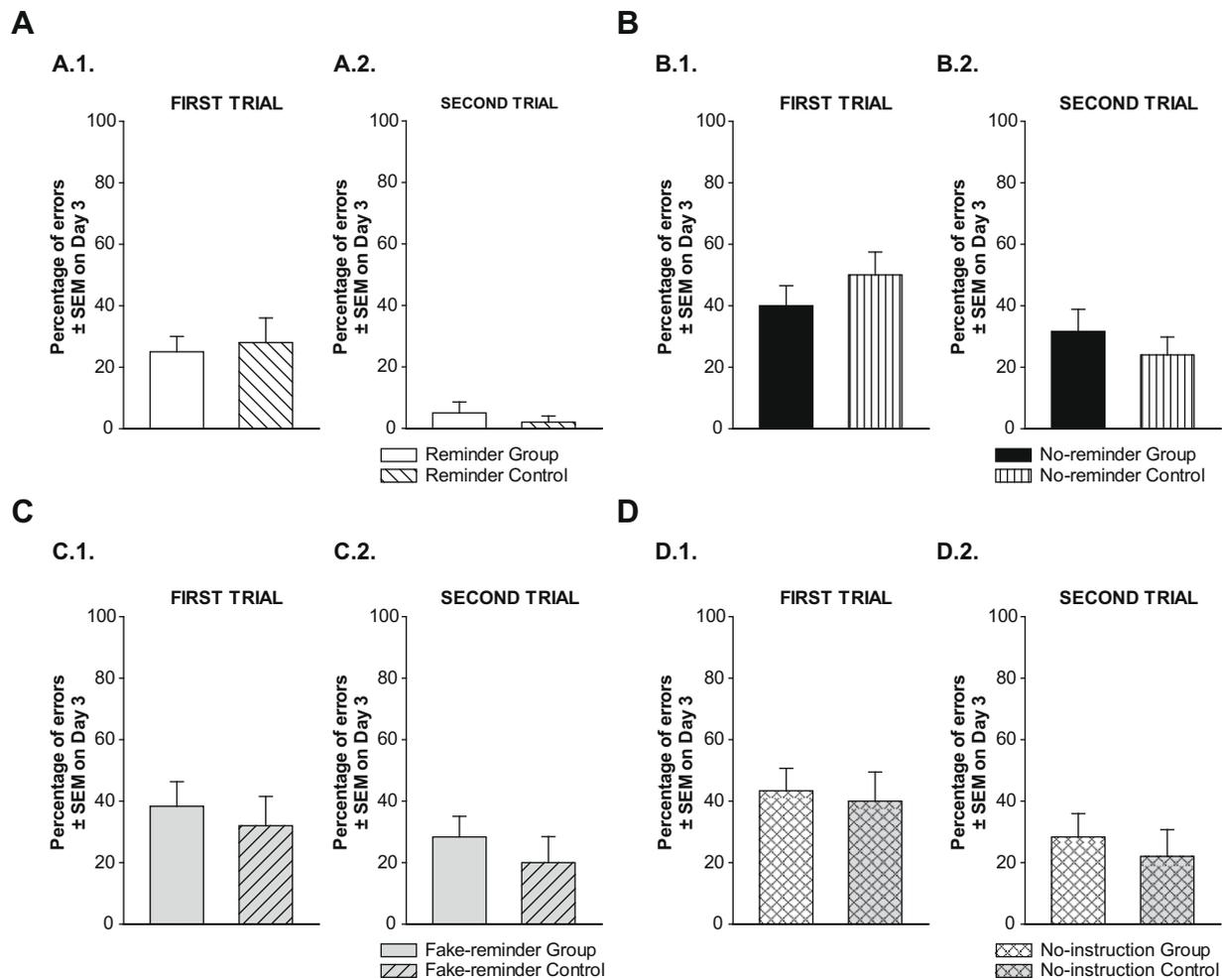


Fig. 4. L1-List control groups analysis. Control Groups ($n = 10$), main groups ($n = 12$) (A) *Reminder-Group* vs. *Reminder-Control*. White bars stand for the *Reminder-Group*, striped bars for *Reminder-Control*. (A.1) First test-trial. (A.2) Second test-trial. (B) *No-reminder-Group* vs. *No-reminder-Control*. Black bars stand for the *Reminder-Group*, striped bars for *No-reminder-Control*. (B.1) First test-trial. (B.2) Second test-trial. (C) *Fake-reminder-Group* vs. *Fake-reminder-Control*. Grey bars stand for the *Fake-reminder-Group*, striped bars for *Fake-reminder-Control*. (C.1) First test-trial. (C.2) Second test-trial. (D) *No-instruction-Group* vs. *No-instruction-Control*. White cross-hatched bars stand for *No-instruction-Group*, grey cross-hatched bars for *No-instruction-Control*. (D.1) First test-trial. (D.2) Second test-trial. All data presented as percentage of errors \pm SEM on Day 3.

the reminder (Pedreira, Perez-Cuesta, & Maldonado, 2004). Moreover, bilateral intrahippocampal infusion of anisomycin in rats, given after reactivation, impairs their ensuing performance provided no information about the location of the new platform is encoded (Morris et al., 2006). Furthermore, the presentation of a second training after L1-retrieval produces amnesic effects in humans on the L1-memory provided the subjects are not allowed to answer with the correspondent response-syllable to the first cue-syllable of the reminder, (Forcato et al., 2009); and here, coincidentally, the same group (now termed *Fake-reminder-Group*) proved to be unsuccessful in triggering labilization-reconsolidation and in permitting L1-memory updating, confirming a direct and strong relation between verbal instruction and reconsolidation.

It was established above that the *INFO-memory*, formed as a new one in the *No-reminder-Group*, the *Fake-reminder-Group* and the *No-instruction-Group*, coexists with its respective previous L1-memory. However, it seems reasonable to suppose that the *INFO-memory* is less strong than the L1-memory, since it is acquired as a new and independent memory in just one trial of only reading the new syllables. This difference would explain two findings in our results. Firstly, when the L1-syllables are tested (Fig. 3B), the recall of the L1-memory in all the main groups, apart from the *Reminder-Group*, recruits items from the *INFO-memory*

that interfere with the former memory, but since the *INFO-memory* is less strong than the L1-memory, a retraining trial is necessary (=the test trial 1) to obtain at test trial 2 a proper interference and a significant difference with the *Reminder-Group*. Secondly, unlike previous papers, (Forcato et al., 2007, 2009) in which the presentation of training like the L1-training after the reminder blocked the L1-memory, no impairment of the L1-memory is shown by *INFO* presentation after the reminder (Fig. 4A), in keeping with the view that *INFO-memory* is less strong than L1-memory.

The present paper deals with the addition of new information into a former memory in the framework of human memory reconsolidation. The possibility that non-factual, but qualifying information, can be added to the memory of an event, out of the reconsolidation process, has been the subject of a consistent line of research for several years. In the foundational paper of Loftus and Palmer (1974), it was shown that if immediately after an event a witness is asked for a precise feature of the event, his recall of such a feature one week later depends on the adjective which was used in the former question to qualify the event. Although the initial thrust of the series was a practical one: to assess how postperceptual (misleading) information could distort an eyewitness report (e.g. Loftus, 1975, 1977, 1979a, 1979b), relevant

theoretical contributions in the field were made later by this research (Loftus & Loftus, 1980; Loftus, 1998; Davis & Loftus, 2006). Thus, Loftus and Loftus (1980) discussed whether the substitution of the memorial information by the new one implies that the former vanishes, which is a topic specially related to the subject of our research. In fact, although in the present paper INFO is incorporated into the L1-memory through addition to the former L1, ongoing experiments in our laboratory are aimed at substituting L1 for INFO and, in such case, the question of the co-existence of both items in the single L1-memory remains to be investigated. Moreover, another influential antecedent concerning our study is the classical research of Izquierdo and Chaves (1988) about the longest time interval at which new information can be added to the initial one, specifically, up to 3 h after training. Here, memory labilization and INFO addition, separated by 5 min, occur 1 day after L1-training, but we offer no information as regards the maximum interval between labilized memory and INFO addition. In our initial paper on memory reconsolidation (Forcato et al., 2007) we reported that after the reminder, the memory continued to be labilized, i.e. susceptible to be disrupted by the interference of new training (L2), up to 6 h. Would the time window for memory disruption be similar to the time window for INFO addition? Further research is required to answer this question.

In brief, the results of this work represent, besides a solid new instance of updating in reconsolidation, a novelty regarding the possibility of using a verbal instruction in order to add new information to a previous L1-memory. Moreover, it is possible to identify the parametrical conditions of the updating in the framework of reconsolidation. Specifically: the reminder needs to be unreinforced and, on the other hand, the verbal instruction needs the order of adding INFO to the previous list L1 of syllables. That is, the sole presentation of the new information is not enough to allow incorporation.

The demonstration that new information enters the labilized former memory entails a critical change in the idea that the memory patrimony of an animal is confined to the memories which are acquired once and which are susceptible to being changed only by forgetting-induced weakening. This patrimony could now be conceived as an assembly continuously enriched not only by the addition of new memories, but also, and mainly, by frequent changes in the content of previous memories. In the case of humans, the scenario is expected to be extremely more dynamic because the new information can quickly enter the labilized memory by using the exclusive tool of language.

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