

Mechanisms of Memory Reorganization during Retrieval of Acquired Behavioral Experience in Chicks: the Effects of Protein Synthesis Inhibition in the Brain

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According to current concepts, memory can be disrupted by administration of protein synthesis inhibitors over a relatively short time period before and after learning. However, data have been obtained indicating that protein synthesis inhibitors can induce amnesia when given long after learning if administration is performed in reminder conditions, i.e., when the animal is presented with one of the environmental components which previously formed the learning situation. The aim of the present work was to confirm the possibility of inducing memory disruption in chicks at late post-learning stages by administering the protein synthesis inhibitor cycloheximide in association with a reminder procedure. Day-old chicks were trained to perform a standard passive avoidance task. Chicks were given cycloheximide (20 µg, intracerebrally) 5 min before the reminder procedure, which was performed 2, 24, or 48 h after training. Testing was conducted 0.5, 1, 3, 24, and 48 h after the reminder. Administration of cycloheximide in association with the reminder procedure induced the development of temporary amnesia, whose duration gradually decreased as the interval between training and reminding increased. These data led to the hypothesis that a memory reactivated by a reminder undergoes a process of reorganization and reconsolidation, which depends on the synthesis of new proteins. The quenching of the ability of protein synthesis inhibition during the reminder to disrupt memory demonstrates the existence of a gradual process resulting in consolidation of memory between 2 and 48 h of learning.

It is now generally accepted that the formation of memory occurs in a number of sequential stages, making up the process of memory consolidation, i.e., the transfer of information acquired during learning from short-term forms of storage into long-term forms [3, 15, 19, 43, 45]. Many of the mechanisms making up the memory consolidation process are regarded as having been deciphered, and the data obtained have been used to propose a cascade scheme of the molecular processes underlying the formation of long-term memory [22, 24, 36, 40]. It has been demonstrated that the molecular mechanisms of memory consolidation in various species of animals have essentially similar features (though they differ in detail); thus, for brevity, we will restrict ourselves to a description of those stages of the consolidation cascade involved in

the passive avoidance model in chicks, which was used in the present studies. The elements of this cascade are activation of synaptic receptors, especially glutamate NMDA receptors [35], followed by activation of second messenger systems, especially Ca^{2+} [12] and cyclic adenosine monophosphate [7], activation of enzymes, especially protein kinases A and C [56, 57], which phosphorylate proteins (transcription factors such as CREB [16]), which in turn trigger the expression of early genes such as the *c-fos* gene [8]; some of the early genes encode effector proteins [6], while other early genes also encode transcription factors such as the *c-Fos* and *c-Jun* proteins [6], which trigger the expression of late genes which encode structural proteins and other molecules required for modifying synapses [42, 43]; in addition, a number of effector proteins undergo glycosylation [32, 42]. The need for these processes for memory formation has been demonstrated in studies using specific inhibitors of each stage [4, 9, 33, 44, 55, and others]. The duration of this cascade

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of molecular processes within cells is believed to be limited to periods of up to several hours after learning, after which memory traces become stable and cannot be disrupted by the blockers of any of the above stages of memory consolidation [5, 22, 40].

However, there are data providing evidence that experimental disruptions to memory can be produced at time points significantly greater than the duration of this cascade mechanism. Thus, studies on rats have demonstrated that electroconvulsive shocks and hypothermia, applied 24 h after training, can disrupt memory when these actions were applied shortly before presentation of one of the components of the learning situation [18, 30, 34]. This procedure of presenting one of the components of learning has been termed reminding; application of electroconvulsive shocks and hypothermia without reminding did not produce this amnesiac effect. Effective reminder procedures include electrical skin stimulation (used as negative reinforcement at the moment of learning) [18], placing the animals in the training chamber for a short period of time, and presenting sounds which made up part of the learning situation [30, 34]. In addition, data have been obtained showing that memory in mice can be disrupted by the protein synthesis inhibitor anisomycin given 3 h after training [25], and that memory in fish is sensitive to disruption by the protein synthesis inhibitor acetoxycycloheximide given 24 h after training [13]. In both cases, the inhibitor induced amnesic effects only on the background of a reminder procedure (consisting of placing the animal in the training apparatus for a short period), but not without application of this procedure [13, 25].

These data suggest that the reminder procedure induces a reorganization of memory during which memory once again becomes sensitive to influences which can disrupt its consolidation. A hypothesis has been proposed, that integration of new information to an already-functioning memory system can occur at the moment of reactivation, this new information requiring its own fixation time [28]; it has also been suggested that reactivation results in the triggering of a second period of memory consolidation [47], i.e., so-called memory reconsolidation [39, 46]. However, the questions of which of the known memory consolidation processes are involved in these "reconsolidation" processes and the extent to which this phenomenon affects different classes of animals remain in need of further study. Thus, the aim of the present work was to study the involvement of one component of the memory consolidation cascade, protein synthesis, in memory reorganization processes occurring in response to a reminder. We selected passive avoidance in chicks [10] as the experimental model, as the sequence of biochemical events underlying memory consolidation has been studied in detail in this model, the durations of each of the stages are known, and data on their disruption by

specific inhibitors have been obtained [31, 36, 37, 40, and others]. In addition, recent studies have yielded data on the possibility of disrupting memory in this model at late time points after training by administration of blockers of short-term memory: sodium glutamate, lanthanum chloride, and the glutamate NMDA receptor blocker AP5 [52–54], when these treatments are given in association with a reminder procedure.

METHODS

Pairs of male Lomon-Brown one-day-old chicks were placed in a metal box of size 20 × 25 × 20 cm. Chicks had free access to standard feed and water throughout the experiment. Illumination was provided by a daylight lamp from 09:00 to 21:00 and the temperature was maintained at 28–30°C. A standard model of one-session training to passive avoidance was used (see [23]). Experiments were performed between 10:00 and 18:00. Chicks were pre-trained at the beginning of the experiments: they were given three opportunities to peck a white bead (2 mm in diameter) fixed to a bar. The interval between presentations of the white bead was 5 min. Because of the innate tendency to peck small, round objects, most of the chicks (85–95%) pecked the bead. Subsequent experiments included only those chicks which pecked the white bead on at least one of the three opportunities. In the training, which took place 5 min after the last presentation of the white bead, chicks were invited to peck a red bead (3.2 mm in diameter) wetted with the caustic substance methylanthranilate (MA). Having pecked this bead, chicks demonstrated typical behavior induced by the caustic taste of MA: they shook their heads, wiped their beaks on the floor, gave out cries, closed their eyes, and cowered down. On testing, i.e., repeated presentation of the red bead, this time dry, most of the chicks (80–100%) avoided it, behavior which in this model indicates an acquired avoidance habit. At the same time, testing using beads of other colors showed that the chicks continued to peck them. Verification of the specificity of the acquired habit in these experiments was performed using a dry white bead, as used for pre-training and (at later testing points), a dry blue bead (3.2 mm in diameter), which had not previously been presented to the chicks. The dry blue bead was used for testing in order to confirm the ability of the chicks to peck a new bead, not associated with the aversive experience, at relatively long periods of time after hatching (more than 48 h) – data have been obtained indicating that spontaneous pecking by chicks of new beads decreases with age [11, 17].

The reminder procedure was performed 2, 24, 48 h after training. The reminder consisted of the test procedure [52], i.e., 10-sec presentations of a dry red bead of

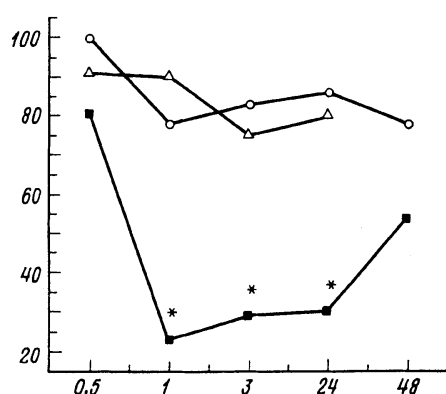


Fig. 1. Disruption of the passive avoidance habit by administration of cycloheximide 2 h after training in conditions of reminding. Testing was at 0.5, 1, 3, 24, and 48 h after the reminder (horizontal axis). Squares show administration of cycloheximide before the reminder ($n = 11, 13, 17, 20$ and 11 respectively for the test time points). Control groups: circles show administration of physiological saline before the reminder ($n = 12, 19, 12, 15$, and 14); triangles show administration of cycloheximide without the reminder ($n = 12, 11, 20$, and 15); the vertical axis shows the proportion of avoidances (%); $p < 0.05$, χ^2 criterion. For explanation see text.

the same size and color as used for training. Chicks were given intracerebral doses of the protein synthesis inhibitor cycloheximide (10 μg per hemisphere) 5 min before the reminder procedure; previous studies have shown that this agent produces amnesia when given 5 min before training when recall is tested 24 and 48 h after training [2, 31]; the injection volume was 5 μl per hemisphere. Cycloheximide was given bilaterally using a microinjector into the intermediate medial ventral hyperstriatum, a brain area playing a critical role in forming memory in the passive avoidance model in chicks [38, 41]. Two types of control group were formed: 1) animals given injections of physiological saline 5 min before the reminder procedure, and 2) animals given cycloheximide injections 1 h 55 min, 23 h 55 min, or 47 h 55 min after training, as in chicks of the experimental group but differing in that they were not subjected to the reminder procedure.

Testing using the red bead was conducted 0.5, 1, 3, 24, and 48 h after reminding, testing using the white bead was performed 15 min after testing with the red bead, and testing using the blue bead was performed 15 min after testing with the white bead. Chicks which did not peck the white bead in these tests were excluded from analysis; the proportion of chicks in this category did not usually exceed 10–15%. Quantitative assessment of reproduction of the habit was based on measurements of the percentage level of avoidance, calculated for each group as the number of chicks avoiding pecking the red bed on testing divided by the total number of chicks which pecked the

red bead during training, multiplied by 100%. Differences between groups were assessed statistically using the χ^2 criterion [1].

RESULTS

The aim of the first series of experiments was to confirm the possibility of disrupting the acquired passive avoidance habit by giving cycloheximide 1 h 55 min after training in conditions of reminder presentation and without the reminder. These experiments used 202 chicks; animals were divided into 14 groups of 11–20 chicks. The results of these experiments are shown in Fig. 1. Chicks given injections of cycloheximide 1 h 55 min after training without use of the reminder showed good reproduction of the habit when tested 0.5, 1, 3, and 24 h after injections (91%, 90%, 75%, and 80% avoidance respectively). Chicks given injections of physiological saline 5 min before reminders also showed good reproduction of the habit when tested 0.5, 1, 3, 24, and 48 h after the reminder (100%, 78%, 83%, 86%, and 78% avoidance respectively). However, chicks given injections of cycloheximide 5 min before the reminder demonstrated good reproduction of the habit only when tested at 0.5 h (81% avoidance) and 48 h (54% avoidance) after the reminder. In the test performed 48 h after the reminder, reproduction was somewhat reduced compared with reproduction in the corresponding control group given injections of physiological saline 5 min before the reminder (78% avoidance), though there was no statistically significant difference. On testing 1, 3, and 24 h after the reminder, reproduction of the habit was significantly disrupted (23%, 29%, and 30% avoidance respectively), and these values were statistically significantly different from the percentage avoidance levels in the corresponding control groups: chicks injected with physiological saline and chicks given injections of cycloheximide without reminders ($p < 0.05$). Thus, injections of cycloheximide given 1 h 55 min after training accompanied by the reminder procedure led to disruption of memory in the time interval 1–24 h after the reminder, but not 0.5 or 48 h after the reminder.

The aim of the second series of experiments was to confirm the possibility of disrupting the acquired passive avoidance habit by injecting cycloheximide 23 h 55 min after training with and without the reminder. These experiments used 185 chicks; the animals were divided into 12 groups of 9–25. The results are shown in Fig. 2. Chicks given injections of cycloheximide 23 h 55 min after training without reminders showed good reproduction of the habit when tested 0.5, 1, 3, and 24 h after injections (93%, 91%, 77% and 83% avoidance respectively). Chicks given injections of physiological saline 5 min before reminders also demonstrated good reproduction of the

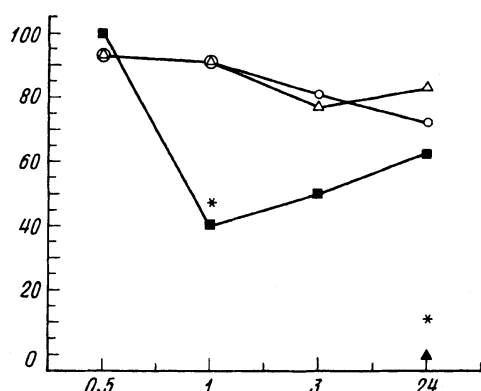


Fig. 2. Disruption of the passive avoidance habit by administration of cycloheximide 24 h after training in conditions of reminding. Testing was at 0.5, 1, 3, and 24 h after the reminder. Squares show administration of cycloheximide before the reminder ($n = 10, 25, 14, 11$). Control groups: circles show administration of physiological saline before the reminder ($n = 15, 24, 14, 11$); light triangles show administration of cycloheximide without the reminder ($n = 15, 23, 9, 12$); dark triangles show testing with the new bead. For explanation see text; axes as in Fig. 1.

habit when tested 0.5, 1, 3, and 24 h after the reminder (93%, 91%, 81%, and 72% respectively). Chicks given injections of cycloheximide 5 min before the reminder demonstrated good reproduction of the habit when tested 0.5 h after the reminder (100% avoidance) and decreased, albeit not significantly in comparison with the control group, reproduction of the habit when tested 3 and 24 h after the reminder (50% and 63% avoidance respectively); however, when tested 1 h after the reminder, reproduction of the habit was disrupted (40% avoidance) to an extent which was statistically significantly different from the level of avoidance in the control groups: chicks injected with physiological saline and chicks injected with cycloheximide without the reminder ($p < 0.05$). Chicks given cycloheximide injections 5 min before the reminder and passing tests 24 h after the reminder were additionally tested with the blue bead. These chicks demonstrated an absence of avoidance (0%), which was statistically significantly different from the level of avoidance of the red bead (63%; $p < 0.05$) and showed that 48 h after training, chicks were perfectly able to peck a new bead but refused to peck the known bead which had previously been associated with aversive properties.

Thus, doses of cycloheximide given 23 h 55 min after training, accompanied by the reminder, led to disruption of memory which was apparent 1 h, but not 0.5, 3, or 24 h after the reminder (see Fig. 2), and, consequently, to shorter-term loss of the habit than that induced by injections of cycloheximide before the reminder, i.e., 2 h after training (see Fig. 1).

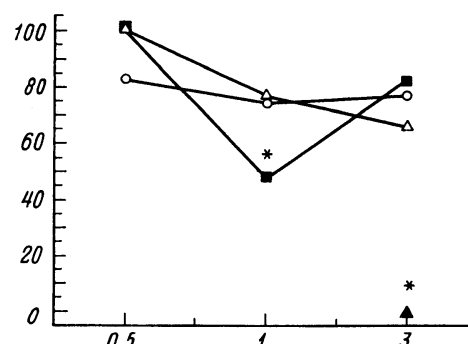


Fig. 3. Disruption of the passive avoidance habit by administration of cycloheximide 48 h after training in reminder conditions. Testing was at 0.5, 1, and 3 h after the reminder. Dark squares show administration of cycloheximide before the reminder ($n = 11, 27, 12$). Control groups: light circles show administration of physiological saline before the reminder ($n = 12, 28, 9$); light triangles show administration of cycloheximide without the reminder ($n = 12, 18, 9$); the dark triangle shows testing with the new bead; the horizontal axis shows time after the reminder (h); the vertical axis shows avoidance (%). For explanation see text.

The aim of the third series of experiments was to confirm the possibility of disrupting the acquired passive avoidance habit by giving cycloheximide 47 h 55 min after training, with and without the reminder. These experiments used 138 chicks, divided into nine groups 9–28 chicks. The results are shown in Fig. 3. Chicks given injections of cycloheximide 47 h 55 min after training without the reminder demonstrated good reproduction of the habit 0.5, 1, and 3 h after cycloheximide doses (100%, 77%, and 66% avoidance respectively). Chicks given injections of physiological saline 5 min before the reminder also demonstrated good reproduction of the habit when tested 0.5, 1, and 3 h after the reminder (83%, 75%, and 77% avoidance respectively). Chicks given injections of cycloheximide 5 min before the reminder demonstrated good reproduction of the habit when tested 0.5 and 3 h after the reminder (100% and 83% avoidance respectively), though when tested 1 h after the reminder, habit reproduction was disrupted (48% avoidance), the difference being statistically significantly different from the percentage avoidance level in control groups consisting of chicks injected with physiological saline and chicks injected with cycloheximide without the reminder ($p < 0.05$). Chicks given injections of cycloheximide 5 min before the reminder and subjected to testing 3 h after the reminder were again tested with the blue bead. These chicks demonstrated no avoidance (0%), which was statistically significantly different from the level of avoidance of the red bead (83%; $p < 0.05$) and was evidence that chicks were perfectly capable of pecking the new bead

51 h after training, but avoided pecking the known bead which had previously had aversive properties.

Thus, injections of cycloheximide given 47 h 55 min after training, accompanied by the reminder, led to memory disruption apparent on testing at 1 h but not at 0.5 or 3 h after the reminder (see Fig. 3) and, consequently, habit loss was even shorter-lasting than after injections of cycloheximide given before the reminder applied 24 h after training (see Fig. 2), as in the latter case the plot showing recovery of habit reproduction was less steep (see the habit reproduction curves in Figs. 2 and 3).

DISCUSSION

The results of the present experiments demonstrate that intracerebral administration of the protein synthesis inhibitor cycloheximide 1 h 55 min, 23 h 55 min, and 47 h 55 min after training to a passive avoidance reflex without the reminder procedure failed to disrupt the acquired habit. This is in complete agreement with current concepts that protein synthesis inhibitors can disrupt acquired behavioral habits only when given 1–1.5 h after training [20, 26, 31, 37, and others]. The generally accepted interpretation of these facts is that protein synthesis is involved in forming memory over a defined period of time immediately after learning [14, 19, 31]. However, our experiments show that intracerebral administration of the protein synthesis inhibitor cycloheximide to chicks in association with a reminder procedure 2, 24, and 48 h after training leads to disruption of subsequent reproduction of the habit. This effect is the result of the action of cycloheximide in association with the reminder procedure, because injection of physiological saline in association with the reminder procedure did not produce the effect. The reminder procedure used here (presentation of a dry red bead which, during training, had a hot, caustic taste) with a significant stimulus for chicks (when they saw the bead at the moment of the reminder, they made anxious sounds, shook their heads, closed their eyes, and wiped their beaks on the floor of the chamber in just the same way as during training). Therefore, the data obtained here, indicating that memory could be disrupted by the protein synthesis inhibitor at this time point, suggest that a biologically significant stimulus (the shape of an object which previously had aversive properties) reactivates a previously consolidated memory in such a way as to cause the memory to undergo consolidation again (to be “reconsolidated”) [39, 46, 48], this process depending on protein synthesis.

It is important to emphasize that the development of amnesia as a result of administration of a protein synthesis inhibitor 5 min before the reminder occurred not immediately, but only 1 h after the reminder procedure

(see Figs. 1–3). Testing 0.5 h after the reminder demonstrated the existence of undisrupted reproduction of the habit (Figs. 1–3). These data are evidence that memory was maintained by some other mechanism during the time interval between the reminder and the appearance of signs of amnesia, this mechanism being independent of protein synthesis.

The dynamics of the development of amnesia associated with administration of cycloheximide before the reminder, seen in our experiments, are similar to the dynamics of the development of amnesia associated with administration of protein synthesis inhibitors before training [20, 55]: when protein synthesis inhibitors were given before training, amnesia developed over time and appeared at 60 min (but not at 30 min) after training using the passive avoidance model in chicks, when cycloheximide [55] or anisomycin [20] was given 5 min before training. These data cannot be explained in terms of the slow onset of action of cycloheximide as a protein synthesis inhibitor: it is known that the level of incorporation of labeled leucine into protein decreases by 74.32% within 30 min of intracerebral doses of cycloheximide to chicks [21]; by 1 h after cycloheximide doses, this measure is still essentially the same – it showed a 75.3% decrease [21], while there are significant behavioral changes (amnesia) at this time as compared with the level of reproduction of the habit 30 min after dosage with cycloheximide [20, 55]. The delayed disruption of habit reproduction is explained on the basis that memory is maintained during the time between training and the appearance of amnesia by mechanisms of short-term and intermediate memory [19, 20]. Consideration of data obtained by Summers et al. on the possibility of disrupting reactivated memory using the short-term memory blocker sodium glutamate (the disruption is detected in tests performed at late periods – up to 24 h after reminding – and, consequently, cannot be explained only by the blockade of the mechanisms of long-term memory) [52] led us to suggest that during memory “reconsolidation,” the initial stage is again one of activation of short-term memory mechanisms (and at this time point, reproduction of the habit is maintained by these mechanisms), which subsequently trigger long-term memory, which is dependent on protein synthesis (and reproduction of the habit disappears in conditions of protein synthesis inhibition).

It follows from our experiments that the sensitivity of memory traces to disruption by protein synthesis inhibitors in conditions of reminding decreases during the interval from 2 to 48 h after training (Figs. 1–3). When memory is disrupted by administration of cycloheximide 1 h 55 min after training (5 min before the reminder), the “time window” during which the habit is significantly disrupted is the interval between 1 and 24 h after the reminder (Fig. 1); when given at 23 h 55 min, the “win-

dow" is narrower – significant disruption occurs at 1 h and there is some decrease in reproduction 3 h after the reminder (Fig. 2); finally, when doses are given at 47 h 55 min, reproduction is disrupted only 1 h after the reminder, while the habit is completely restored by 3 h after the reminder (Fig. 3).

These results are in good agreement with data obtained by Summers et al. [52–54], which demonstrated that the period during which memory can be disrupted by administration of short-term memory blockers in conditions of reminding was quite long, amounting to about 48 h after training. Summers et al. [51] advanced the hypothesis that completion of memory consolidation in the passive avoidance task in chicks needs about 48 h and not the 1–1.5 h suggested previously, because of the "time window" during which protein synthesis inhibitors are effective when given without the reminder [14, 20, 31, 37, and others].

In addition, Summers et al., like ourselves (this report), found that the possibility of disrupting already-formed memories in the passive avoidance model in chicks using sodium glutamate and AP5 in reminder conditions also decreased over the time period between 7.5 min and 48 h after training [52, 54]. These data suggest that the proposed process of memory consolidation, occurring at up to 48 h after training, is gradual in nature.

Which biochemical and molecular processes underlie the proposed consolidation of memory in the passive avoidance task in chicks during the 48 h after training currently remains unknown. However, it is important to note that in the absence of the reminder procedure, as our data showed, administration of cycloheximide 1 h 55 min, 23 h 55 min, and 47 h 55 min, and, as shown by Summers et al., administration of sodium glutamate, lanthanum chloride, and AP5 7.5, 40 and 120 min and 24 and 48 h after training [52–54] did not disrupt these processes.

Such a long period of memory consolidation (up to 48 h after training) is not in accord with current concepts of the duration of molecular-genetic processes underlying long-term memory [6, 20, 22, 40]. However, studies of memory consolidation processes in mammals have demonstrated that there are two forms of consolidation [6]. The first of these is associated with the molecular cascade and is restricted to the few hours after training. Concepts of the second form of consolidation were developed from studies of hippocampus-dependent declarative memory in mammals; consolidation in this case refers to transfer from the short-term memory store, the hippocampus, into the permanent store, the neocortex [51]. This transfer can take prolonged periods of time: about 1–2 weeks in rodents [27, 50], 2–12 weeks in monkeys [58], and several (sometimes up to 25) years in humans [49, 51] (the exact time depends on the learning conditions and the type of action used to detect the possibility

of disrupting memory). Abel et al. termed this prolonged consolidation "late transformation" to discriminate between consolidation at the molecular level and consolidation associated with the transfer of memory traces from the hippocampus to the neocortex [3]. Our data suggest that birds may also have the second type of memory consolidation, similar to the prolonged consolidation of declarative memory in mammals.

Thus, these published data led us to suggest that decreases in the effectiveness of the reminder procedure over the period from 2 to 48 h after training is associated with the fact that reactivation of memory is accompanied by its "reconsolidation" only when the memory trace is relatively new. In the case of our learning model, the period of "relative novelty" of memory traces is no greater than 48 h after training.

It follows from our data that disruption of the habit due to administration of cycloheximide in association with a reminder 2, 24, and 48 h after training is temporary – the habit recovers spontaneously by 3–48 h after the reminder (Figs. 1–3).

Recovery following disruption of an already-formed memory has also been demonstrated previously: thus, disruption of a formed habit in mice using the protein synthesis inhibitor anisomycin, given 3 h after training on the background of a reminder, showed that memory recovered spontaneously by three days after training [25]. A habit disrupted in rats by hypothermia applied 24 h after training on a background of a reminder was restored by the following day [29, 30]. In addition, studies involving the disruption of an already-formed passive avoidance habit in chicks using sodium glutamate and the NMDA receptor antagonist AP5 in conditions of a reminder also yielded data showing spontaneous recovery of memory by 24–48 h after the reminder [52, 54]. Use of lanthanum chloride as the amnesia-inducing agent resulted in even earlier recovery of the disrupted habit – by 10–15 min after the reminder [53].

Previous studies have shown that memories disrupted in chicks by administration of a protein synthesis inhibitor during training did not recover spontaneously [2, 31, 37]. It is also known that amnesia induced in rats and mice with protein synthesis inhibitors given during training lasts several days and even a week after training [14]. It is therefore apparent that the nature of memory disruption by protein synthesis inhibitors is different when these agents are given during training and reminding. Judge and Quartermain [25] suggested that the phenomenon of spontaneous recovery was associated with the hypothesis that not all the neurons activated during training are activated during the reminder procedure; rather, only a proportion of these neurons are activated, and this part of the neural network and only this part is subject to disruption of memory. Thus, the effects of

amnesia-inducing agents on "old" memories must be less effective than their effects on "new" memories [25]. Mac-tutus et al. [29] put forward another explanation, based on the suggestion that during reminding, the initial memory trace store is intact, while exposure to an amnesia-inducing agent changes only the "retrievable properties." Subsequently, after repeated tests, access to the memory can become increasingly possible because of the undisrupted initial memory store [29]. A similar point of view has been suggested by Summers et al. [52–54] – that the gradual recovery of memory disrupted by administration of sodium glutamate on a background of reminding might be evidence that the fundamental memory trace is not activated directly during retrieval and is inaccessible to disruption [52]. However, the hypothesis of Judge and Quartermain [25] does not explain the basis on which the "significantly disrupted" habit, which is not detected on testing, can be restored, while the explanation proposed by Summers et al. throws no light on how, if the fundamental memory trace remains intact, we could detect amnesia on testing. Thus, data obtained by ourselves and other authors on the spontaneous recovery of habits which had been disrupted long periods of time after training raise a question which is poorly explained from the points of view of existing concepts of the mechanisms maintaining memory at times when the memory is not evident in behavior and the mechanisms underlying spontaneous recovery and leading to the possibility of retrieving the memory after it has been in such a "latent state."

The fact that similar data on the disruption of already-formed memories on the background of a reminder procedure have been obtained previously in studies of other classes of animals – mice [25] and goldfish [13] – in other models of learning, and in chicks using the same model of learning [52–54] but using different amnesia-inducing agents, is evidence that the phenomenon described above reflects some common fundamental mechanisms which operate during the reorganization of long-term memory.

CONCLUSIONS

1. Presentation of a biologically significant component of learning (a reminder) 2, 24, and 48 h after training leads to reorganization ("reconsolidation") of memory, which is dependent on protein synthesis.

2. The decrease in the possibility of disrupting memory by protein synthesis inhibitors given on the background of a reminder demonstrates the existence of a gradual process of memory consolidation occurring between 2 and 48 h after training. These data suggest that birds may have the second type of memory consolidation (the so-called late transformation), which is similar to the

prolonged consolidation of declarative memory in mammals.

3. Memories disrupted by administration of cycloheximide in conditions of reminding 2, 24, and 48 h after training recovered with time. The data obtained here raise the question of the possible mechanisms of this recovery, as well as the question of the mechanisms maintaining memory in the "latent form" during the time period in which it is not evident in behavior.

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