RESEARCH ARTICLE

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Long-term retention of motor skill in macaque monkeys and humans

Abstract Remarkable human performance, such as playing the violin, is often based on motor skills that, once acquired, are retained for a long time. To examine how motor skills are retained, we trained monkeys and humans extensively to perform many visuomotor sequences and examined their performance after a long retention period of up to 18 months. For both monkeys and humans, we found strong evidence for long-term retention of motor skills. Each of the monkey subjects initially learned 6–18 sequences of button presses extensively by trial-and-error for up to 18 months. After a long retention period, they were asked to perform the previously learned (OLD) sequences together with completely new (NEW) sequences. The performance for OLD sequences was much better than for NEW sequences in terms of accuracy (assessed by the number of errors to criterion) and speed (assessed by the performance time). However, the retention was interfered with in two conditions, but in selective manners: (1) Learning of other sequences during the retention period interfered with accuracy, but not speed, of performance; (2) Inter-manual transfer was absent for speed, but not accuracy, of performance. The human subjects performed basically the same task as the monkeys. Each subject initially learned one sequence of 20 button presses by trial-and-error during an 8–10 day learning session. After 16 months, they were asked to perform the previously learned sequence (OLD sequence) and additional sequences including RECENT sequences (learned one day before) and NEW sequences. Their performance was considerably better on OLD and RECENT sequences than NEW sequences. Whereas the number of errors (reflecting 'accuracy') was lower for RECENT than for OLD sequences, the performance time (reflecting 'speed') was shorter for OLD than for RECENT sequences. Interestingly, the subjects were unaware that they had experienced OLD sequences. The results suggest that a motor skill is acquired and retained in two different forms, accuracy and speed. This occurs separately but concurrently. This conclusion is consistent with the hypothesis that at least two neural mechanisms operate independently to represent a motor skill.

Keywords Procedural memory · Sequence task · Interference · Inter-manual transfer

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Introduction

Various kinds of motor skills, such as typing, juggling, piano playing, pursuit rotor task, and gymnastic skills, can be retained for a long time (Hovland 1951; Annett 1979; Arthur et al. 1998). However, many questions remain to be examined. For example, what kind of information is retained and what is lost? Does practice induce changes in the nature of memory representation? These questions are particularly important for studying the neural mechanisms of motor skills. Although recent functional imaging studies have shown that many areas in the human brain are related to acquisition of motor skills (Salmon and Butters 1995; Doyon 1997; Willingham 1998; Hikosaka et al. 1999a), few studies have been aimed at examining long-term retention of skills.

Motor skill is obviously not unique to the humans. Non-human primates and other mammals can acquire complex motor skills (Iwaniuk and Whishaw 2000). However, most studies on long-term skill retention have been conducted on human subjects. Although behavioral neuroscientists are aware of the animals' ability of developing motor skills, few studies have been done to investigate it using a rigorous method. This situation is problematic because physiological studies on experimental animals are crucial for understanding the neural mechanisms of motor skills. It has been demonstrated that extensive practice of a motor task leads to structural changes of single neurons (Klintsova and Greenough 1999). However, the process leading to the end result is unclear.

In this context, it would be ideal to study both the human and animal subjects using the same task. We thus trained monkeys and humans on a sequential button-press task extensively to acquire motor skills, and then examined long-term retention of these skills under different conditions. The task, which we call 2×5 task (Hikosaka et al. 1995), was originally devised for monkey subjects and was then applied to human subjects as 2×10 task. These tasks have been used for behavioral and physiological experiments (Hikosaka et al. 1999b).

A prominent feature of the 2×5 and 2×10 tasks was that as many new sequences as required can be generated. All sequences had the same rule and equivalent difficulties. The degree of retention was examined by comparing the performances between the previously learned procedures and the new procedures. The studies to date have trained each subject on only one or a few kinds of procedure (Annett 1979; Arthur et al. 1998), and therefore can only examine the retention by comparing the subject's performance on separate sessions separated by more than 1 year, which could be confounded by non-specific performance changes, such as changes in general motor or cognitive abilities due to laboratory experience, development, and aging.

We found that both monkeys and humans acquired sequential motor procedures in similar manners and, furthermore, retained the motor skill for a long time, more than 1 year. We also found that two aspects of motor skill, accuracy and speed, are expressed, modified, and transferred differently during or after a long-term retention period.

Material and methods

Experimental animals

We used three male Japanese monkeys (*Macaca fuscata*): monkeys P (7.7 kg), M (8.0 kg), and G (5.5 kg). The monkeys were kept in individual primate cages in an air-conditioned room where food was always available. At the beginning of each experimental session, they were carried to the experimental room in a primate chair. The monkeys were given restricted amount of fluid during experiments. Their health condition, including factors such as body weight and appetite, were checked daily. Supplementary water and fruit were provided daily. The experiments were carried out while the monkeys' heads were fixed and their eye movements were recorded. For this purpose, a head-holder and an eye-coil were implanted during surgical procedures (Hikosaka et al. 1995). All surgical and experimental protocols were approved by the Juntendo University Animal Care and Use Committee and are in accordance with the National Institutes of Health Guide for the Care and Use of Animals.

Human subjects

Four subjects (three male and one female) participated in this experiment. They were students of Juntendo University whose ages were 22–23 years at the time of the retention test. All subjects signed a written consent prior to participation.

Apparatus and behavioral paradigm

The monkey and human subjects (simply designated as subjects hereafter, unless otherwise stated) were trained to perform a sequential button-pressing task, called the 2×5 task (for monkeys) and 2×10 task (for humans) (see Fig. 1A). Details were described in a previous paper (Hikosaka et al. 1995). In front of the subjects, who were seated on a chair, was placed a panel on which 16 light-emitting diode (LED) buttons were mounted in a 4×4 matrix. The LED buttons were square in shape (20 mm × 20 mm for monkeys; 12 mm × 12 mm for humans), separated from each other (center-to-center) by 40 mm for monkeys and 20 mm for humans. At the bottom of the panel was another LED button which was used as a home key. To have the monkey use only one hand for button-press, a vertical Plexiglas plate was attached to the chair between the panel and the hand not being used. To change the operating hand, the plate was shifted to the other side. The monkey's head was fixed by a head-holder connected to the primate chair. The monkeys used either the thumb or index finger to press the buttons. Each monkey used the same finger consistently throughout experiment. Human subjects were asked to use one finger (index or middle finger) of their dominant hand to press the buttons.

At the start of a trial, the home key was turned on. When the subject pressed the home key for 500 ms, 2 of the 16 target LEDs turned on simultaneously; this pair is called a 'set'. The subject had to press the illuminated buttons in the correct order, which he/she had to discover by trial-and-error. If successful, these LEDs turned off as they were pressed and another pair of LEDs, a second set, was illuminated, which the monkey had to press again in a correct order. A total of five sets (for monkeys) or ten sets (for humans) was presented in a fixed order for completion of a trial; these were called a 'hyperset'. When the subject pressed a wrong button, all LED buttons were illuminated briefly with (for monkeys) or without (for humans) an unpleasant beep sound, and the trial was aborted without any reward. The subject then had to start over again

from the home key to begin a new trial. A trial was designated as successful only when the subject completed the whole hyperset. The same hyperset was repeated until the subject completed the hyperset successfully for a total of 20 trials (called a 'block'). The monkey was given a liquid reward after each successful set. The amount of the reward increased gradually from the first to the final set so that it was maximized by completing all sets. Human subjects were given no reward and were simply asked to perform the task as quickly and as accurately as possible.

A major advantage of the 2×5 and 2×10 tasks was that as many new hypersets as possible can be generated practically (Hikosaka et al. 1995). Each time a new hyperset was introduced, the subject learned it as a new visuo-motor procedure while retaining the ability to perform hypersets that had already been learned (Hikosaka et al. 1995). In this way, each subject could acquire a repertoire of many learned hypersets.

Initial learning

Monkeys

During the initial learning period, the monkey subjects performed 6–18 hypersets on every experimental day, one block each day, over a long time (approximately from 3 to 18 months) until the hypersets could be performed very skillfully as a repertoire of well-learned procedures or motor skills. The monkey was required to use the right hand for half of the hypersets and the left hand for the other half, consistently for each hyperset. In addition, the monkeys performed many hypersets that were learned only once (one block). After this initial learning period, the practice for the learned hypersets was stopped for 6–18 months (retention period). During the retention period, the monkeys stopped performing the 2×5 task completely, except for one experiment (interference condition) for monkey P, in which the monkey continued to perform other learned and new hypersets (see Fig. 3A). Monkey G was trained on saccade tasks that were unrelated to the 2×5 task and performed it during the retention period.

Humans

During the initial learning session, each subject learned one 2×10 hyperset for 8–10 blocks, one block each day. In addition, the subjects performed many hypersets for other experimental purposes including two hypersets that were learned only once (one block).

Retention test

Monkeys

The retention test was conducted as a 1-day session after the retention period. The monkeys performed the previously learned hypersets (called OLD hypersets) and new hypersets (NEW hypersets), and continuously learned hypersets (LEARNED) in the interference condition for monkey P. The order of these types of hypersets was counterbalanced. The monkeys used the same hand as that used for practice, except for one experiment (transfer test) for monkey G in which half of the previously learned hypersets (including OLD hypersets) were performed with the hand opposite to the practicing hand (see Fig. 4). One-half of the new hypersets was performed with the right hand, the other half with the left hand.

Humans

The test experiment was conducted 16 months later as a 2-day session (see Fig. 7). On day 1, the subjects learned two new hypersets. On day 2, the subjects performed seven hypersets, which included the previously learned hypersets (OLD) in addition to two NEW hypersets, two RECENT hypersets (the hypersets learned on day 1), and two ONCE hypersets (the hypersets experienced only once 16 months previously). After the performance of each hyperset, the subjects were asked whether they had performed the hyperset before with a five-grade rating (1 no, 2 probably no, 3 undecided, 4 probably yes, 5 yes). The order of the sequences was counterbalanced across the subjects.

Data analysis

We used two parameters to assess the accuracy and speed of performance. As a measure of 'accuracy', we counted the number of error trials before completing one block (20 successful trials). Only sequence errors that occurred when the subject pressed the two illuminated buttons in the wrong order were included. To evaluate 'speed' we measured a mean performance time, which was the time from the home key-press to the second button-press of the final set, averaged across 20 successful trials. All statistical analyses were done by using the software package STATISTICA. The null hypothesis for retention was that the number of errors or the mean performance time was the same between OLD hypersets and NEW hypersets. This hypothesis was tested using a t-test when only OLD and NEW hypersets were compared. When various types of hypersets were compared overall in human experiments (OLD, ONCE, RECENT, and NEW), a mean value across the two hypersets for each subject was obtained for each of ONCE, RECENT and NEW conditions. Based on these mean values and a value obtained from an OLD hyperset, we performed a one-factor ANOVA with repeated measures and a post-hoc Newman-Keuls test among various types of hypersets.

Results

Long-term retention in monkey subjects

Evidence for long-term retention of motor skills

Figure 1 shows the basic schedule for the initial practice and the retention test based on our strategy. During the initial learning period, the monkeys learned many hypersets (n=6–18) over a long time (3–18 months), always using the same hand for a given hyperset: half of them with the right hand, the other half with the left. The monkeys also performed several hypersets every experimental day as new hypersets. These experiments overall were done for behavioral analysis of procedural learning (Hikosaka et al. 1995; Miyashita et al. 1996; Rand et al. 1998, 2000) and for physiological experiments (Miyachi et al. 1997; Lu et al. 1998; Nakamura et al. 1998, 1999).

Two behavioral measures, the number of errors and the mean performance time, were obtained for each daily experiment (one block of 20 successful trials for each hyperset). The number of errors decreased rapidly, while the performance time decreased gradually (Fig. 1B), consistent with a previous report (Hikosaka et al. 1995). During the daily sessions of about 20 days, the monkeys' performance changed from a reactive mode to an anticipatory mode, such that both the eyes and hand moved to the target position before it was illuminated (Miyashita et al. 1996). After an extended practice period of more than 3 months or so, the monkeys could perform the learned hypersets very quickly with few errors, which
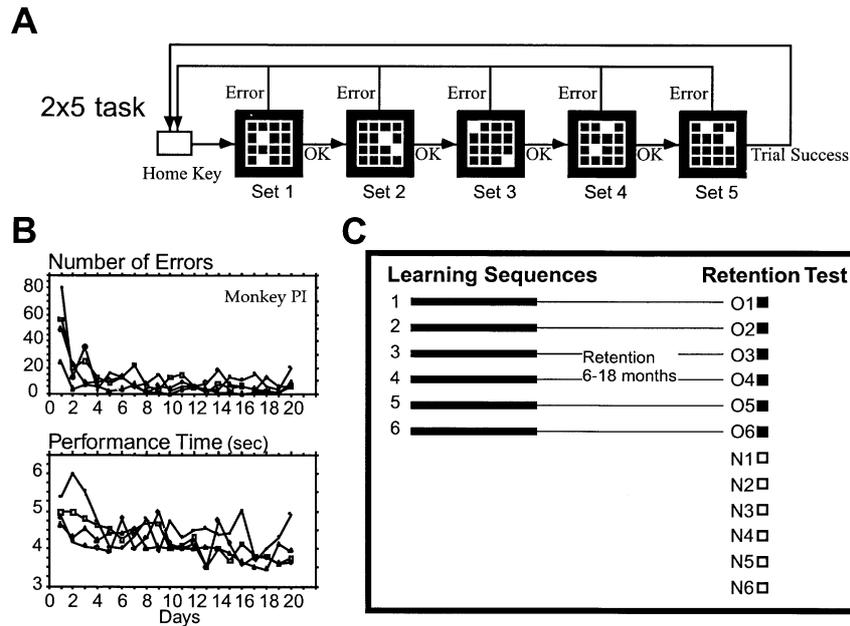
Fig. 1A–C Long-term retention test of motor skill in monkeys. **A** Procedure of the 2×5 sequence task. The monkey had to press the two illuminated buttons in the correct order to proceed to the next set until completing five sets (called a 'hyperset'). Each hyperset is presented repeatedly in a block until the monkey performed the whole hyperset for a total of 20 trials. A different hyperset was then used for the next block. **B** The learning curves of monkey P for four hypersets during the initial practice period of 20 days are shown as the number of errors before completing 20 trials successfully (*top*) and the mean performance time for the successful trials (*bottom*). **C** General schedule of the long-term retention test. After retention periods of 6–18 months, the monkey was asked to perform the previously learned hypersets (*O1–O6*) and new hypersets (*N1–N6*) in a 1-day session. Data obtained in monkey subjects are shown in Fig. 2, 3, 4, 5, 6
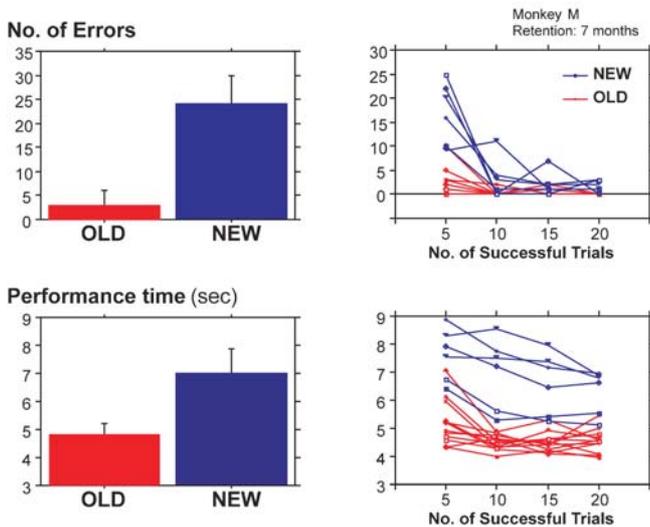

Fig. 2 Evidence for long-term retention of motor skill in monkeys. After a retention period of 7 months, monkey M performed 12 OLD hypersets and 6 NEW hypersets. Bargraphs indicate the number of errors and the mean performance time for OLD and NEW hypersets (means +SE). Plots indicate the changes in the monkey's performance within a block of 20 successful trials for individual OLD and NEW hypersets. The within-block performance was divided into four parts based on the number of successful trials (1–5, 6–10, 11–15, 16–20), and the two parameter values were calculated for each part

might be called visuomotor skills (Hikosaka et al. 1995; Lu et al. 1998).

The practice was then stopped altogether for a set of the learned hypersets (*n*=6–18), and was followed by a long-term retention period (6–18 months) (Fig. 1C). After the retention periods, the monkeys were asked to perform the previously learned hypersets (OLD hypersets, indicated as O1-O6). On the same day, the monkeys also performed a similar number of new hypersets (NEW hypersets, N1-N6) for comparison.

Figure 2 demonstrates the long-term retention of motor skill. After a retention period of 7 months, monkey M performed 12 OLD hypersets, each using the same hand as that used for previous practice (right six, left six). In addition, he performed six NEW hypersets (right three, left three). Both the number of errors and the performance time were significantly lower for OLD hypersets than for NEW hypersets [number or errors $t_{(16)}=10.03$, $P<0.0001$; performance time $t_{(16)}=7.09$, $P<0.0001$]. As shown in the plots of Fig. 2 the monkey made many errors for NEW hypersets during the initial quarter of one block (until five successful trials). In striking contrast, the number of errors for OLD hypersets was close to zero from the beginning. The performance times for NEW hypersets decreased during one block of practice, but hardly reached those for OLD hypersets, even their initial values. Note that the monkey experienced no hyperset during the retention period.
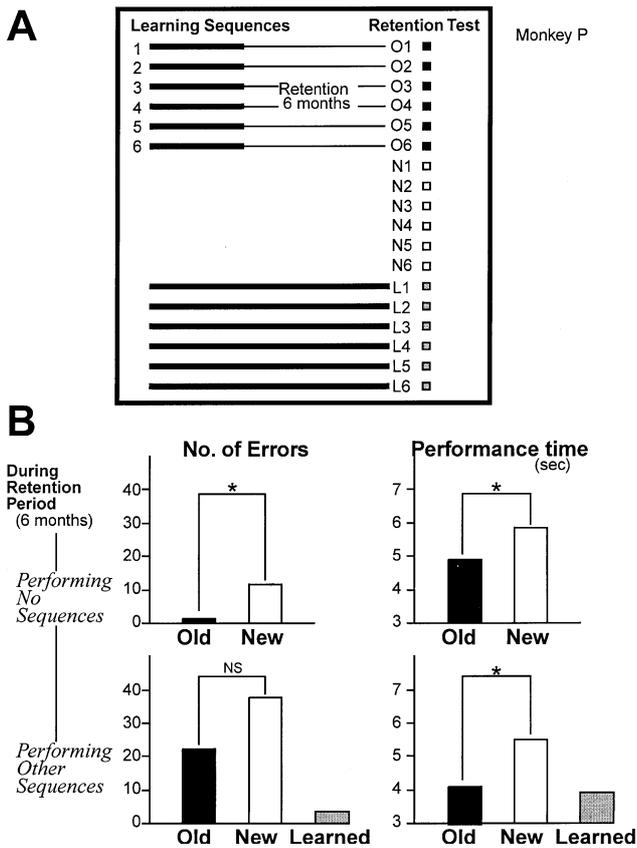
Fig. 3A,B Effects of interference on motor skill in monkeys. **A** Schedule for the retention test in the interference condition. During the retention period (6 months), monkey P performed the previously learned hypersets (*O1–O6*) and new hypersets (*N1–N6*) and continued to perform 14 other learned hypersets (*L1–L6*) and learned many new hypersets (not indicated) (interference condition). In the non-interference condition, monkey P performed no hyperset during the retention period. **B** The results of the retention test for the no-interference (*top*) and interference (*bottom*) conditions, shown for previously learned (OLD), new (NEW), and continuously learned (LEARNED) hypersets. The numbers of hypersets were: 14 OLD and 13 NEW for the no-interference condition, and 6 OLD, 6 NEW, and 14 LEARNED for the interference condition. The results of statistical comparison between OLD and NEW hypersets are indicated by the significance levels: *NS* not significant; *$P<0.01$ (t-test)

Selective interference between motor skills

A distinguishing feature of our 2×5 or 2×10 task was that each subject can learn many sequential procedures based on the same rule. We asked whether the memories for these procedures interfere with each other. For this purpose, we examined monkey P under two different schedules (no-interference condition and interference condition), but with the same 6 months of retention period (Fig. 3). Figure 3A shows the schedule for the interference condition. The test for the interference condition was performed first. Monkey P then learned other hypersets for 11 months and then the test for the non-interference condition was conducted.
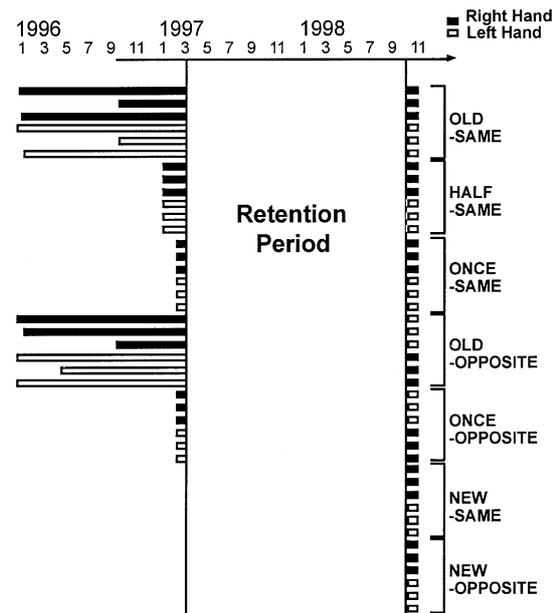

Fig. 4 Experimental schedule of 'transfer' test. Monkey G learned 12 hypersets (six with the right hand, six with the left hand) for 6–15 months (OLD) and six hypersets (three right and three left) for 3 months (HALF). The monkey experienced no hyperset during the retention period of 18 month. On the day of the retention test, the monkey performed 42 hypersets: OLD and HALF hypersets (*n*=18), 12 hypersets that the monkey performed only once (one block) on the day before the retention period (ONCE), and 12 new hypersets (NEW). Among OLD and ONCE hypersets (*n*=24), half was performed with the hand used for practice (SAME) and the other half with the opposite hand (OPPOSITE); HALF hypersets were performed only with the same hand. Half of 12 new hypersets were randomly assigned to the same hand (NEW-SAME) and the other half to the opposite hand (NEW-OPPOSITE)

In the no-interference condition, which was the same as that used for monkey M (Fig. 2), the monkey performed no hyperset during the retention period. The results (Fig. 3B *top*) were qualitatively the same as those for monkey M (Fig. 2): both the number of errors and the performance time were significantly lower for OLD than for NEW hypersets [number or errors $t_{(25)}$=5.33, $P<0.0001$; performance time $t_{(25)}$=3.82, $P<0.001$].

In the interference condition, the monkey continued to perform six other learned (LEARNED) hypersets (Fig. 3A). The results (Fig. 3B *bottom*) were different from those for the no-interference condition in several ways. Firstly, the number of errors was not significantly different between OLD and NEW hypersets [$t_{(10)}$=0.53, $P>0.05$], while the performance time was shorter for OLD hypersets [$t_{(10)}$=4.59, $P<0.01$]. Secondly, the number of errors was higher while the performance time was shorter in the interference condition than in the no-interference condition, both for OLD hypersets [number of errors $t_{(18)}$=11.11, $P<0.0001$; performance time $t_{(18)}$=2.82, $P<0.05$] and NEW hypersets [number of errors $t_{(17)}$=5.17, $P<0.0001$; performance time $t_{(17)}$=3.48, $P<0.01$]. The comparison between OLD and LEARNED hypersets was interesting: the number of errors was much
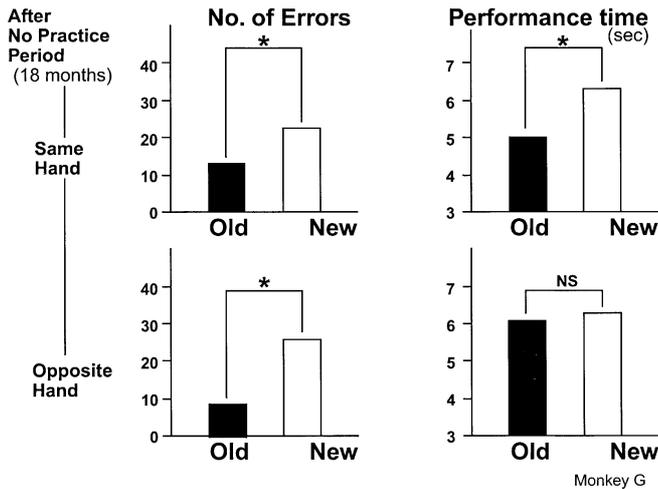
Fig. 5 Results of 'transfer' test: OLD-NEW comparison in monkey G. The number of errors was significantly lower for OLD hypersets than for NEW hypersets either using the same or opposite hand (*$P<0.05$, t-test). The performance time was significantly shorter for OLD hypersets when using the same hand (*$P<0.01$, t-test), but not when using the opposite hand (*NS*, $P>0.05$, t-test)
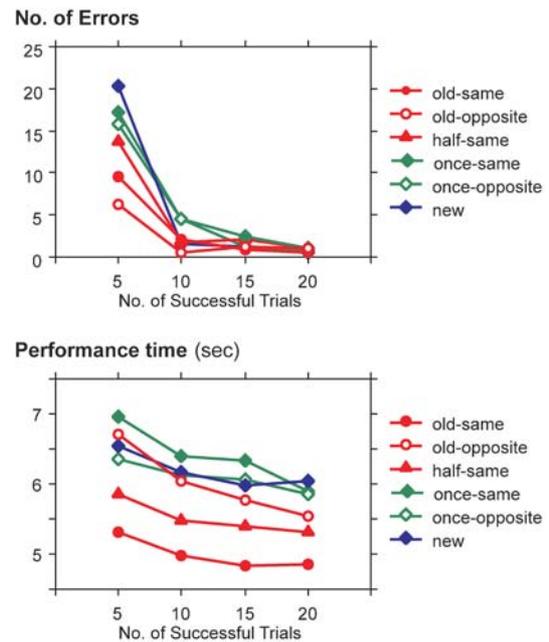

Fig. 6 The changes in the monkey's performance within a block of 20 successful trials for differently scheduled hypersets (transfer test, Monkey G). The performance for ONCE hypersets was similar to that for NEW hypersets. The retention of the performance time, not the number of errors, was specific to the hand used for practice, while both parameters were dependent on the duration of the practice period. Condition designations as in Fig. 4

lower for LEARNED hypersets [$t_{(18)}$=7.64, $P<0.0001$], whereas the performance time was only weakly lower for LEARNED hypersets [$t_{(18)}$=2.35, $P<0.05$].

These results suggest that acquisition of different motor skills interferes with retention of old motor skills as well as acquisition of new skills in terms of accuracy (represented by the number of errors), but not speed (represented by the performance time). Instead, the speed seems to be improved in general by experiencing new and different sequences.

Selective transfer of motor skills

We now asked whether a motor skill learned with one hand transfers to the opposite hand. As shown in Fig. 4, monkey G initially learned many hypersets extensively. Depending on the duration of practice, they were grouped into OLD (>6 months) and HALF (3 months) hypersets. In addition, the monkey experienced many hypersets for only one block (ONCE). The monkey used the right hand consistently for half of the hypersets and the left hand for the other half. After a long retention period of 18 months (during which the monkey was engaged in saccade tasks), the retention test was performed on 1 day with different combinations of hands. A main question was how the monkey performed OLD or HALF hypersets using the hand opposite to that used for practice, after such a long-term retention period.

The results for OLD hypersets again showed differential effects on accuracy and speed (Fig. 5). For OLD hypersets compared with NEW, the number of errors was significantly lower regardless of the hand used [same hand $t_{(10)}$=2.59, $P<0.05$; opposite hand $t_{(10)}$=3.49, $P<0.01$]; in contrast, the performance time was signifi-

cantly shorter when the same hand was used [$t_{(10)}$=4.61, $P<0.001$] but was not significantly different when the opposite hand was used [$t_{(10)}$=0.60, $P>0.05$].

The results of all combinations are shown in Fig. 6 as the change in performance within a block of trials. The monkey made most errors before completing five successful trials. Errors occurred mostly in the first quarter (before completing five successful trials), roughly depending on the duration of the initial practice in the increasing order of OLD, HALF, ONCE, and NEW. A similar tendency was present for the performance time as long as the same hand was used: the order from OLD-SAME, HALF-SAME, to ONCE-SAME was consistent throughout the block. However, the results were completely different when the opposite hand was used: the performance time in OLD-OPPOSITE condition was virtually indistinguishable from ONCE-OPPOSITE or NEW, in the first two quarters, although it started decreasing in the later half of the block.

These results suggest that motor skill is stored in two forms, the memory for accurate performance, which is accessible also to the unpracticed hand, and the memory for speedy performance, which is limited to the practiced hand.
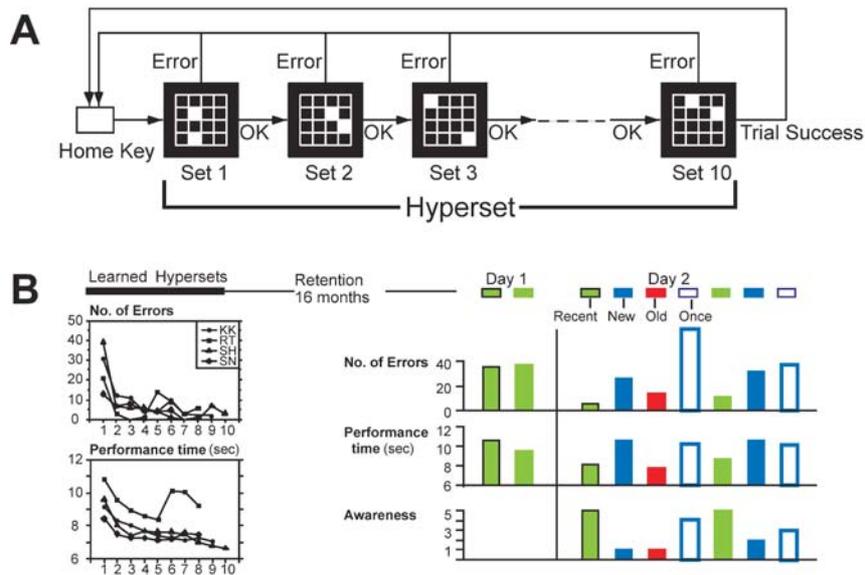
Fig. 7A,B Test for the long-term retention of motor skill in human subjects. **A** Procedure of 2×10 task (see Materials and methods section). **B** Experimental procedure of the retention test. During the initial learning (*left*), each subject learned one 2×10 hyperset for 8–10 day sessions. The learning curves for each subject are shown for the number of errors before completing 20 trials successfully (*top*) and the mean performance time for the successful trials (*bottom*). After the retention interval of 16 months, the subjects underwent the test experiment over 2 days (*right*). The performance (number of errors and performance time) of one subject SH is shown: *Day 1* two new hypersets; *Day 2* the hyperset that he learned extensively during the initial learning session (OLD), two hypersets that he had learned on day 1 (RECENT), two new hypersets (NEW), and two hypersets that he performed only for one block during the initial learning session (ONCE). On day 2, the subject rated the awareness for each hyperset from 1 ('I am sure I have never done it') to 5 ('I am sure I have done it'). Further data obtained in human subjects are shown in Figs. 8 and 9

Long-term retention in human subjects

We found that our human subjects learned a 2×10 hyperset (Fig. 7A) similarly to monkeys (Fig. 7B, for comparison with Fig. 1B). During the initial learning session, the number of errors decreased quickly, already reaching a minimum level by day 2 or 3. The performance time decreased more gradually until the end of the session, except for subject RT. In addition to the particular hyperset to be learned, each subject learned several 2×10 hypersets for different experimental purposes (subject KK 12, RT 8, SH 2, SN 5) and experienced many hypersets only once. During the retention period of 16 months, the subjects had no experience on the 2×10 task and received no warning of the test experiment.

The test experiment was done as a 2-day session. The performance of subject SH is shown in Fig. 7B. On day 1, the subject performed two new 2×10 hypersets with many errors and long performance times. On day 2, the subject performed seven 2×10 hypersets: the hyperset that he learned extensively during the initial session (OLD), two hypersets that he had learned on day 1 (RECENT), two new hypersets (NEW), and two hypersets that he performed for only one block during the initial learning session (ONCE). The number of errors for OLD hyperset was lower than for two NEW hypersets and for two ONCE hypersets, but was higher than for two RECENT hypersets. Interestingly, however, the performance time was shortest for OLD hyperset. On day 2, we asked the subject, after each hyperset, whether he had ever performed the hyperset. This subject was confident in indicating that he experienced RECENT hypersets while he had not experienced OLD hyperset. The answers for NEW hypersets and ONCE hypersets were inconsistent.

Similar results were obtained in the other subjects (Fig. 8). As group data, the subjects' performance and awareness were significantly different among NEW, ONCE, RECENT, and OLD hypersets [number of errors $F_{(3,9)}=12.25$, $P<0.01$; performance time $F_{(3,9)}=33.31$, $P<0.0001$; awareness $F_{(3,9)}=17.67$, $P<0.001$]. Both the number of errors and the performance time were smaller for the learned (RECENT or OLD) hypersets than for NEW hypersets (post-hoc Newman-Keuls test, $P<0.05$), indicating that the motor skill was retained for as well for 16 months as for 1 day. Among the learned (RECENT and OLD) hypersets, the number of errors was lower for RECENT hypersets and the performance time was shorter for OLD hyperset (post-hoc Newman-Keuls test, $P<0.05$). These results suggest that the speed of performance, rather than its accuracy, is retained for a long time, more than 1 year.

Subjects SN, KK and SH were very similar for these comparisons. Subject RT was somewhat different in that her performance on OLD hyperset was poorer than the other subjects in terms of both the number of errors and the performance time; the performance time was even slightly longer for OLD hyperset than the RECENT hypersets. This might be related to the atypical learning process of this subject: the performance time of subject
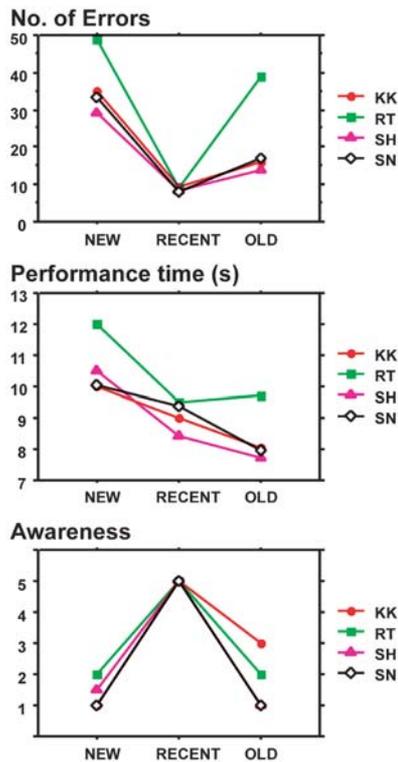
Fig. 8 Comparison between the NEW, RECENT, and OLD-learned hypersets for the performance (number of errors and performance time) and awareness of the four human subjects. The data values for the NEW or RECENT hypersets are the average of the two hypersets examined

Fig. 9 Within-block changes of the number of errors (*top*) and the performance time (*bottom*) in human subjects. The within-block performance was separated into four parts based on the number of successful trials (1–5, 6–10, 11–15, 16–20), and the two parameter values were calculated for each part. The data are the average of the four subjects, shown separately for four sets of hypersets: (1) new hypersets that were used on the second day of the test session (NEW); (2) hypersets that were used twice in the test session over day 1 (NEW-CTRL) and day 2 (RECENT); (3) hypersets that were used only once in the initial learning session (ONCE); (4) hypersets that were learned extensively in the initial learning session, the data shown for the first day of learning (NEW-ORIG) and the last day of learning (LEARN-ORIG), and tested 16 months later (OLD)

RT decreased until day 5, but then increased suddenly on day 6–8 (Fig. 7B).

This observation suggested that the performance level at the end of the initial learning period determined the degree of long-term retention (see Annett 1979). To test this hypothesis, we compared the number of errors and the performance time between the last day of the initial learning session and the day of the retention test. There was significant correlation for the performance time (regression analysis $r=0.98$, $P<0.05$), but not for the number of errors ($P>0.1$). This is unlikely to be due to inter-individual differences because no correlation was found for ONCE hypersets ($P>0.05$).

Despite the speedy performance for OLD hyperset, no subject had clear memory that he/she had experienced the hyperset before (Fig. 8 *bottom*). While all subjects remembered RECENT hypersets, the subjects' awareness of OLD hypersets was very low and was not statistically different from that of NEW hypersets (post-hoc Newman-Keuls test, $P=0.52$). Taking together RECENT and OLD hypersets, there was a significant correlation between the awareness score and the number of errors (regression analysis, $P<0.05$), but no significant correlation between the awareness score and the performance time ($P=0.08$). The results suggest that the accuracy of performance, rather than its speed, is correlated with awareness.
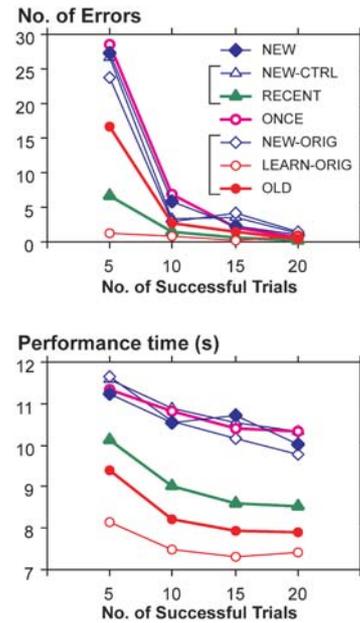
The difference of the subjects' performance on different types of learned hypersets (RECENT and OLD) was clear and consistent throughout a block of trials to reach the criterion of 20 successful trials (Fig. 9). The subjects made most errors before completing five successful trials. Nonetheless, the mean number of errors was consistently lower for RECENT hypersets than for OLD (Fig. 9 *top*). In contrast, the mean performance time was consistently shorter for OLD hypersets than for RECENT.

Figure 9 also shows other significant results. The performance on OLD hypersets was worse than the performance on the same hypersets just before the retention period (LEARN-ORIG), indicating that 'forgetting' occurred for both parameters (Arthur et al. 1998). The performance on ONCE hypersets was virtually identical with that for NEW, indicating that learning only once had no effect 16 months later. Note the subjects experienced new hypersets in three different contexts: NEW, used on the second day of the test session; NEW-CTRL, the same as RECENT but tested on the first day of the test session; NEW-ORIG, the same as OLD but tested on the first day of the initial learning session. They were very similar to each other, supporting our postulate that the average performance on new 2×10 hypersets was

stable and any deviation from it toward a better performance indicates the retention of memory.

These results suggest that long-term retention of motor skills was expressed more strongly in speed than in accuracy, whereas newly acquired motor performance was expressed more strongly in accuracy than in speed. Newly acquired motor performance involved awareness, but not long-retained motor skills.

Discussion

Methodological issues for assessment of long-term retention

Serious care must be taken to assess long-term retention of motor skills. Firstly, during the retention period, the subjects must have no chance to perform the test skill or even an action similar to the skill. This requirement for a test procedure precludes motor skills that could be performed in daily life. There is probably no such possibility for the performance on the 2×5 or 2×10 task. Even so, the retention test can be done only once after a long-retention period because the retention test itself is inevitably part of practice.

The last point leads to a second difficulty: control of readiness and motivation of the subjects at the time of the retention test. This is particularly problematic when we use animal subjects. Even if the subjects are well motivated to work for reward, they may have difficulty in handling the sudden encounter with the previously familiar apparatus and environment. Many of the previous results may have been confounded by this type of error because retention was assessed by comparing the originally learned performance and the retained performance. Our retention test was devoid of this type of error, because we compared the performance on previously learned procedures and that on new procedures which were performed within the retention test session.

However, it was difficult to collect data from many subjects with a long retention period in between. This was particularly true for animal subjects because it took a long time to train them to acquire motor skills and then the animals were prohibited to perform the skills during the long retention period. Consequently, our animal research was based on a collection of single-subject studies. This leaves the possibility that the conclusions drawn from our data are constrained to the particular subjects used in this study.

Acquisition and retention of motor skills in monkeys and humans

We found that monkeys and humans retained motor or procedural skills for a long time. What skills were retained was not a basic rule for performing a task, but multiple motor skills based on the same rule. The meaning of this distinction may be clearer by the following analogy: expertise in piano playing for a professional pianist (basic rule), and a repertoire of music pieces for a professional pianist (multiple motor skills based on the same rule). The distinction was unclear in many of the previous studies that examined retention of learned performance for a single motor task, such as pursuit rotor task (Annett 1979). Particularly remarkable in our study was that even monkeys retained not only a general rule of the 2×5 task but also individual motor sequences.

In both humans and monkeys, retention of motor skills was indicated by fewer errors (reflecting accuracy) and shorter performance time (reflecting speed) for previously learned sequences than for new sequences. Interestingly, these parameters varied depending on the context, such as the presence of interference and a change in the motor effector. In the following, we discuss each of the context-dependency.

Interference of memory

Interference between different sets of learning is an old issue in human psychology which has been investigated for more than a century (Hovland 1951). Some studies have been done also on macaque monkeys (Jitsumori et al. 1988). It has a practical impact because efficient learning would then be dependent on subsequent learning of a different item. However, most of these studies dealt with learning of declarative knowledge and therefore it is still unclear whether the same interference occurs for procedural or motor learning. We found that interference indeed occurs also for motor skill learning, but in different ways for the two aspects of learning: accuracy and speed.

One monkey was tested for long-term retention in two different contexts: during the retention period, the monkey continued to perform other learned sequences (interference condition) or not (no-interference condition). In the interference condition compared with the no-interference condition, the performance on average was less accurate (more errors), but was quicker (shorter performance times) (Fig. 3). Another interesting comparison for the interference condition was between the OLD sequences and the continuously learned sequences (LEARNED sequences). The results indicated that the performance for the OLD sequences in the interference condition was similar to that for NEW sequences in terms of accuracy and was similar to that for LEARNED sequences in terms of speed. These results suggest that only performance accuracy, not performance speed is affected by learning of additional sequences. These different effects of LEARNED sequences on two parameters suggest that at least two separate brain mechanisms are involved in long-term retention, one for accuracy and the other for speed.

Why then was there such a biased effect of interference for accuracy and speed? A common interpretation is that the consolidation of the memory for the original

learning continues after learning and therefore can be interfered with by additional learning (see Hovland 1951). Our results might then suggest that the memory for accuracy is less well consolidated than the memory for speed. This is consistent with our data on human subjects, which showed that the memory for accuracy (measure as the number of errors) was worse for OLD sequences than for RECENT sequences whereas the memory for speed (measured as the performance time) was better for OLD sequences than for RECENT sequences.

Effector specificity of memory

Another important issue in learning and memory is transfer. During the initial learning, monkeys used the right hand for half of the sequences and the left hand for the other half. That is, the monkey used the same hand throughout the initial learning of a given sequence. After a long retention period (18 months) monkey G was asked to use the same (learned) hand for half of the OLD sequences and the opposite (naive) hand for the other half. Compared with NEW sequences, the performance accuracy for the OLD sequences was better by using either hand, but the performance speed for the OLD sequences was better in the same-hand condition, but not in the opposite-hand condition. These results suggest that the memory for speedy performance, but not for accurate performance, is specific to the hand used for the initial learning.

The results are consistent with previous studies from our laboratory showing that the accuracy memory is more transferable than the speed memory (Rand et al. 1998). When monkeys were asked to use the hand opposite to that used for practice, the number of errors was lower for LEARNED sequences than for NEW sequences, whereas the performance speed for LEARNED sequences was not different. However, since these tests were done at a period during long-term practice, they did not reveal the ultimate nature of long-term memory. The present study now indicates that long-term memory of a visuo-motor sequence consists of accuracy memory and speed memory and that accuracy memory, not speed memory, is transferable between motor effectors (i.e., hands). Our current and previous studies together suggest that the differentiation of the accuracy memory and the speed memory occurred during memory encoding, not during the retention period.

Multiple neural mechanisms for motor skill

These results suggest that procedural memory or motor skill is stored in at least two forms, one responsible for accurate performance and the other for speedy performance. This model might be called an 'accuracy-speed dual memory model'. With practice, the memory for accuracy would be created earlier than the memory for speed. However, once the memory for speed is created

with long-term practice, it is more robust than the memory for accuracy in that it survives after a long time of no practice. Awareness of performance would be associated with the accuracy memory, not the speed memory.

Relevant to the dual memory model, recent studies have suggested that motor skill learning may be the integrative product of multiple neural mechanisms, each contributing to a different aspect of learning (Shadmehr and Brashers-Krug 1997; Doyon et al. 1998; Willingham 1998; Krakauer et al. 1999). With practice, accuracy of performance was acquired earlier than speed (Hikosaka et al. 1995). Skill learning was associated with dynamic changes in human cortical activation (Toni et al. 1998; Petersen et al. 1998). The accuracy tended to be effector-unspecific while the speed was effector-specific (Rand et al. 1998, 2000; Bapi et al. 2000). A change in motor effector affects activation of sensorimotor cortex, but not parietal cortex (Grafton et al. 1998). Awareness of performance is correlated with activation of prefrontal cortical areas, but not sensorimotor cortex (Honda et al. 1998). The motor cortex may be equipped with dual mechanisms for fast and slow learning (Karni et al. 1998).

Physiological experiments on monkeys and functional magnetic resonance imaging experiments on human subjects suggested that different brain areas are related to the accuracy memory and the speed memory. Thus, frontal cortical areas, especially the presupplementary motor area (pre-SMA; Nakamura et al. 1998, 1999) and the dorsolateral prefrontal cortex (Sakai et al. 1998, 1999) together with the anterior part of the basal ganglia (Miyachi et al. 1997, 2002), contribute to learning of new sequences (corresponding to RECENT sequences in this study), suggesting that these areas are mainly related to the accuracy memory. In contrast, the middle part of the putamen (Miyachi et al. 1997) and the cerebellar dentate nucleus (Lu et al. 1998) contribute to the performance of well-learned sequences(corresponding to OLD sequences), suggesting that these areas mainly represent the speed memory. We have proposed that a parallel neural network model in which a sequential motor skill is represented in two different coordinates, the visuo-spatial coordinates and the motor coordinates (Hikosaka et al. 1999a), and demonstrated its superior performance compared with a single network model (Nakahara et al. 2001). The present study, together with the network model, suggests that a motor skill is retained as a long-term memory mainly in the cortico-basal-ganglia-cerebellar system using the motor coordinates, which supports a speedy, rather than accurate, performance.

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