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Motor memory consolidation in children: The role of awareness and sleep on offline general and sequence-specific learning

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Abstract

Study aim: The purpose of this study was to investigate the role of sleep and awareness on consolidation of general and Sequence-Specific learning in children.
Material and methods: Male participants (n = 48, 10 to 12 years old) were assigned to one of four groups based on awareness and sleep. Acquisition phase took place in the morning (wake groups, 8 ± am) or in the evening (sleep groups, 8 ± pm) followed by a 12 hours retention interval and a subsequent delayed retention test (1 week). Children in the explicit groups were informed about the presence of the sequence, while in the implicit groups were not informed about it. For data analysis in consolidation of general sequence learning and Sequence-Specific Consolidation phases, 2 × 2 × 2 and 2 × 2 × 3 ANOVA with repeated measures on block tests were used respectively.
Results: The data provides evidence of offline enhancement of general motor learning after 12 hours which was dependent on sleep and awareness. Moreover, the information persistence after 1-week was significant only in sleep groups. The results also indicated that consolidation of sequence-specific learning was only observed after 12 hours in element duration and it was related to sleep and awareness.
Conclusions: The results revealed that sleep wasn't only an essential factor in enhancement of off-line sequence learning task after 12 hours in children, but performance of the children was dependent on awareness and sleep.

Keywords: Consolidation – Children – Offline learning – Passage of time – Explicit knowledge

Introduction

Memory consolidation refers to the stabilization and enhancement processes that can occur without any additional practice, intent, and/or awareness [24]. This process can result in increased resistance to interference and an improvement in performance following an offline period [26]. Therefore, memory consolidation involves two phases: stabilization (behavioral performance maintenance) and enhancement (also known as offline learning). Offline learning commonly occurs after sleep without any training or experience [2, 17, 35]. In fact, in this phase, a novel and initially unstable task representation is strengthened and stabilized in long term memory via sleep [9]. This finding is consistent with active system consolidation theory and synaptic homeostasis hypothesis [4, 6, 36]. These theories suggest that a passive process such as sleep can provide optimal conditions for an effective and active enhancement of memory and stable memory representation [1, 4, 6, 10].

Special attention has been given to the role of sleep in learning and consolidation of motor skills, in particular, motor sequential skills [2, 5, 13,]. Motor sequential skills are a type of procedural memory which are learned through the repetition and practice of a sequential pattern without the person's attention to the learning. These skills are a fundamental part in our learned motor repertoire, ranging from simple to complex skills [29, 41].

Some studies demonstrated that performance of motor sequence tasks were improved when the offline period included being a sleep rather than awake, while encoding and retrieval of memories took place preferentially during waking [1, 2, 10, 18, 20, 41]. Many of these studies have concluded that sleep can promote integration of newly acquired information into existing memory schemas and as a result, it allows for better recall of information [18].

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As a matter of fact, sleep has an important role in consolidation of sequential motor skills from the childhood to adulthood. Hence, many researchers have confirmed the benefit of the sleep in both declarative memory (verbalize knowledge of facts and events, in which information recall is conscious) and procedural memory (skills memory) in adults [2, 11, 15, 20, 27, 38]. Although motor learning is an especially critical factor during childhood, sleep is essential in children's cognitive, behavioral, emotional, and motor development and important links to memory and cognition [16, 41]. However, remarkably little is known about the influence of sleep on motor memory processes and learning especially in motor sequence learning during childhood which is still unspecified [7, 35, 41].

Some researches expressed that both children and adolescence's performance improve in declarative memory consolidation after one night sleep. However, this improvement was not sleep related in procedural memory [7, 16, 31, 32, 41]. Other researchers found that sleep can be useful both in children and adults' declarative and procedural memory consolidation after training for a new motor skill [3, 41]. Some of these results contradict the observed findings in researches involving adult participants [7, 16, 32, 35, 41]. Some researchers such as Jongbloed-Pereboom et al. [21] discovered that offline learning was independent of age, while Fischer et al. [16] showed this factor was connected to age.

Accordingly, sleep-dependent motor memory consolidation in children is not well-specified. The underlying reasons for such inconsistency in results are still unknown. It seems that benefits of sleep in enhancement of offline motor sequence learning depends on different factors. One of these factors is the individual's awareness of the task regularities [2, 20, 33, 34, 37]. This may play an important role in children's motor learning and may also be an important factor in offline enhancement. Learning new skills with practice can be accomplished unintentionally, with little to no awareness (implicit knowledge), or intentionally, with an individual's conscious awareness of the regularities of the task to be learned or rules and facts on how to move (explicit knowledge) [2, 21, 33, 37, 42]. Lots of evidence exists about the role of awareness in online and offline motor learning of adults [2, 11, 12, 33, 34, 38].

Some researchers believed that the process of the explicit knowledge in working memory and storing of declarative knowledge in the first stages of learning were essential parts in the performance and learning of the motor skills. On the other hand, some others expressed that lack of explicit knowledge around the motor's basics and regularities did not have a negative impact on the learning process, and even in physiological and psychological stress, it could improve the performance. The results indicated the benefits of the implicit learning along with explicit learning in skills acquisition (online learning) [21, 37].

In offline learning domains, Robertson et al. [33] were the one of the first to demonstrate that awareness of task regularities impacts offline motor learning. Subsequently they presented awareness theory and also sleep-dependent memory consolidation theory [33, 34]. They proposed that when motor sequence learning was implicit, offline enhancement only occurred in the passage of time. However, in explicit conditions, improvement in performance only occurred when the participants experienced sleep especially in the first 12 hours after acquisition phase [2, 34, 38]. These theories have been confirmed with adult's findings by a variety of tasks [2, 20, 21, 33, 38].

Sugawara et al. [35] suggested that sleep is associated with offline skill enhancement in explicit motor sequence task in children, as in adults. Peiffer et al. [31] showed overnight gains of declarative (explicit) memory retention performance in children. However, other researchers showed that sleep, compared to wake in retention test, enhanced the consolidation of implicit motor sequence tasks. These findings were in contrast with the studies which stated that sleep was only beneficial in explicit skill learning [3, 11, 15].

It is noteworthy that relatively few studies have evaluated implicit and explicit learning in children. Van Abswoude et al. [37] investigated the capacity of working memory in implicit and explicit learning in children. They found that after the offline period, enhancement only occurred in accuracy (not speed) in both implicit and explicit learning conditions and minimal differences existed between explicit and implicit learning in children [37]. However, there is lack of evidence in children. Most studies in this field have been conducted on adults which manifested the contradictory evidence on the role of sleep in implicit sequence learning [2, 12, 20, 33]. Some researchers proposed that implicit and explicit learning and their consolidation in adults is different from children [37].

However, researchers, investigating procedural and declarative memory in children, used simple tasks which were naturally implicit (i.e., serial reaction time, implicit continuous task) or explicit (i.e., two-dimensional objects location, word-pair associates, finger tapping task) compared to adults [3, 41]. However, implicit and explicit knowledge of task instructions and regulations were typically not manipulated in these sequence tasks researches [3, 7, 13, 16, 31, 41]. So, considering the little evidence in children and not having enough investigations of the type of knowledge relating to the task sequence, we cannot generalize the results of the adult researches to the children.

On the other hand, most of the studies investigating consolidation in regard to children, evaluated the general improvement of the task related to sequence learning [24]. General skill learning refers to increasing speed and accuracy as a result of practice with the task, though regarding children, there exists another type of learning and consolidation following it, namely sequence – specific learning.

Sequence-specific learning refers to acquisition of sequence-specific knowledge, which results in relatively faster responses for events that can be predicted from the sequence structure versus those that cannot (such as new or random sequence) [24, 26, 37].

The vast majority of experiment on sequence learning have not reported any differences between the general and sequence-specific learning especially in children. Only a limited number of experiments have focused on this issue. Research on young adults and older adults with respect to implicit sequence learning conditions generally revealed that enhancement of general sequence learning had occurred. However, regarding sequence-specific learning, no improvement was found in either age group (young and elderly adults) or consolidation intervals (12, 24-hr & 1 week) [24, 25]. Also, the only research that was done on children showed that improvement only occurred in terms of accuracy (not speed) of general and sequence-specific learning after one day [37]. However, in this field, there exists lack of evidence.

Moreover, most of the previous researchers investigated the children's consolidation process after acquisition phase in a limited time [3, 7, 37, 41]. It is possible that the little persistence of memories after sleep prevented the beneficial effects on the performance in a relatively short period of time after acquisition phase, but after passage of the additional time, performance enhancement might be observed. Though, it is still not clarified whether the overnight gains are temporary or stable after passage of the time in children.

Based on the above discussions, sleep dependent consolidation and its related effective factors like implicit and explicit knowledge are rarely investigated. In fact, the role of awareness in promoting motor sequence learning little is known about whether explicit knowledge of sequence enhances online and offline motor sequence learning in children. Some researchers believe that implicit learning is independent of age and cognitive resources. Consequently, it has been recommend as superior to explicit learning, especially for children [21, 37].

By investigating these factors, we can investigate the theories and hypothesis related to the consolidation and awareness of children in studying the effects of night sleep immediately after the first practice of motor sequential skill. The separation of the enhancement of the general skill from the enhancement of specific sequence in offline and online learning was not adequately addressed in the literature.

Therefore, the purpose of the present research was to evaluate the effect of children's explicit and implicit knowledge in a sleep-dependent consolidation of general and sequence specific motor sequence task. This task involves dynamic arm movement task. It is important to note that detecting fixed sequence within this type of task is more difficult than with a finger tapping and types of serial reaction time tasks [8, 30]. Most of the tasks used in children field were simple and needed little motor requirements (Press the key/button with fingers such as serial reaction time task, finger tapping task, and button box task). The task in the present study, due to the higher level of processing requirements (containing more elements and goals) and more motor needs (flexion/extension movements) were more complex than the previous motor sequence tasks [8, 29, 30]. Therefore, memorizing the information and detecting sequence blocks were more difficult for the participants [30].

Finally, the effects of the night sleep after skill acquisition in long term has little been investigated. Therefore, this article has investigated not only the role of night sleep immediately after the practice of motor sequence skill, but it has also investigated the evaluation of the results of night sleep with the passage of time after one week for determining the resistance of dynamic arm movement task.

Material and methods

Participants

Right hand dominate children (N = 60, age, range: 10–12 years, mean: 10.84 ± 0.72) took part in the study. Informed parental and child consents were obtained. All children were asked to respond to a General Health Questioner (Lndygraf and Abaz, 1996) and reported good general health with no medical conditions, no history of neurological and developmental disorders, no prior experience to the task, no recognized sleep problems and all had normal IQ [3, 14]. They received a small gift for their participation. The experiment was approved by ethics committee of biological research of Ferdowsi University of Mashhad (IR.MUM.FUM.REC.1397.11). Participants were asked to respond to a hand dominance questionnaire [28], Child Sleep Habit Questionnaire and Wechsler Intelligence Scale for Children-Fourth Edition [14, 40]. Some of the participants were omitted from the study: three of which because of lack of sleep, four, of guessing the sequence in the implicit groups, three, of absence in the following 1 week test, one, of absence in retention of 12 hours and one of them omitted because of fatigue and doing the sequence falsely. Finally, 48 children were chosen.

Apparatus

The apparatus was Dynamic Arm Movement Task (DAMT) which was adapted from Park and Shea task [30] to evaluate motor sequence learning. DAMT consist of horizontal lever and monitor (43 inches). The axle of lever which rotated freely in ball-bearing supports, allowed the lever to move in the horizontal plane over the table surface. At the distal end of the lever, a vertical handle was attached. The position of the handle could be adjusted so that when the participant rested their forearm on the lever, with their elbow aligned over the axis of rotation and they could comfortably grasp the handle (palm vertical). The location of the participants' hand on the lever was adjustable to their hands' length [8, 29]. The horizontal movement of the lever was monitored (1000 Hz) by increment rotary encoder, which was attached to the end of the axle of lever and stored for later analysis on computer. A pointer was attached to the end of the lever extended, so that it could be positioned within the targets on the monitor. Also, to reduce the noise, nine optical sensors were used on the main body of the apparatus under the lever to precisely elaborate the movement. Another pointer was attached vertically under the lever to make connection with optical sensors. The distance between the pointer and the monitor was 20 cm and the distance between the participants and the monitor was approximately 80 cm [22, 30].

Procedures

Participants were randomly assigned to one of four experimental groups which differed in terms of the time of day for acquisition testing (sleep and wake) and awareness (implicit and explicit) of the sequence. The sleep groups started acquisition phase at 8 PM (±1 hour) and the wake groups started it at 8 AM (±1 hour) [41]. In the explicit groups the participants were provided knowledge about the order of the sequence elements and what trials/blocks this sequence would be presented, but the implicit groups were not informed of the repeating fixed sequence [33, 37]. The wake groups were not allowed to take a nap, but the sleep groups were instructed to sleep after the acquisition session [13, 24, 41].

The participants were seated on a chair facing the monitor and the apparatus was adjusted, so that the participants' lower arm was approximately on the 60-degree angle to the upper arm at the starting position which arbitrarily designated as 0-degrees. The range of the motion required to complete the sequences was approximately 0 to 80 degrees from the start position. The participants had to move the pointer to the targets displayed on monitor by flexion and extension of their arms [22]. The diameter of the targets represented 2 degrees of elbow extension/flexion. The targets (10 circles) were illuminated on the monitor but only four of the targets (1, 4, 7, and 10) were actually used in the sequence. To begin a trial, participants were asked to move the lever to the start position. When the home position was achieved, outlines of the targets were illuminated and the first target 20 degrees from start position (Target 1) was illuminated. Targets 2–10 were spaced at 6.67 degree increments. Before the acquisition phase, participants completed one random block (R) in order to get familiar with the task. Upon hitting the target (crossing the boundaries of target circle) the illumination was turned off and the next target was immediately illuminated until the block was completed. If the participants missed a target, the target remained illuminated until the participant returned the lever to the target position. After hitting the last target in each block, a ''stop'' tone was presented and the display of the targets was removed [22, 30]. Participants were instructed to respond as quickly, smoothly, and accurately as possible to the changing target location that appeared on the screen by moving the lever from one target to next [8, 22].

The acquisition phase included 10 training blocks with 96 targets in each block (12 elements × 8 repeat) with a one minute rest between each block [24] .The order in which the targets were illuminated in S[1]$_2$ – S_4, and S_6 – S_8 and S_{10} was based on the predetermined pattern. In Blocks R_1, R_5 and R_9 the targets were illuminated in a random order. The predetermined order of the goal sequence was 4,1,4,1,4,7,4,1,4,7,10,7 with the same distance between each target (20 degrees). The same movement distances were used in the random blocks but the target pattern was changed [30]. To limit the participant's ability to acquire knowledge of the sequence the first three elements of each block were randomly presented and these elements were omitted from the analysis [24].

Retention test were conducted approximately 12 hours after the completion of the acquisition session. Follow up test took place at the same time of day (Between 08:00–10:00 AM) after one week for all groups [13, 24]. Each test included three blocks (96 trials). The first and third blocks (S_{11} and S_{13} in the retention test and S_{14} and S_{16} in the follow-up test) involved the same sequence as used on repeated blocks in acquisition phase and the second block (R_{12} in the retention test and R_{15} in the follow-up test) included a random sequence [13]. The experimental design is shown in Appendix.1. Sleep duration and its quality after acquisition phase were evaluated from parents and children's verbal reports [41].

Finally, at the end of the test, explicit knowledge of sequence was examined by a brief interview with the participants. They were asked if they had noticed anything in particular about the stimulus locations and responses [17]. They were asked to report the order of the repeated sequence, either by guessing or from memory. The training sequence was judged to have been acquired if after the experiment the participants correctly reported 6 or more elements of the 12 elements training sequence [24]. In addition, a recognition test was conducted with the participants seated in front of the monitor and keyboard. Five different demo sequences were shown with one of the sequences being the same as the one that they learned in the

[1] Sequence Block.

acquisition phase of the experiment. If they answered correctly, we concluded that they had acquired explicit and declarative knowledge of the repeated sequence [30]. This knowledge required conscious processing, and it was different from the processing method of implicit groups. This was because the requirement for implicit learning was non-conscious processing of the sequence orders such that participant did not aware the pattern and order of sequences during the practice and recognition test [33, 34].

Data analysis

For data processing MATLAB software (Math works, R2014a) was utilized and for statistical analysis SPSS 22 was used. Experimental variables included element duration and error of prediction. Element duration was computed as elapsed time from hitting (crossing the target boundary) of the currently illuminated target to hitting the next illuminated target. Error of prediction was indicated when a reversal movement was made away from the intended target in a sequence. As a result, when an unnecessary reversal was observed in the movement displacement while reaching to the goal, we concluded that the participants had misjudged the next target in the sequence.

Analysis of the data was performed for several different phases that exist in this experiment with their corresponding blocks. General improvement was determined by analyzing changes in performance across the acquisition phase. Sequence-specific learning was determined using the mean difference of element duration and error of prediction between R_9 and the average of S_8 and S_{10} (R_9 – Mean (S_8, S_{10})). Consolidation of general learning was determined by comparing the last block of acquisition phase (S_{10}) and the first block of the 12-hour retention test (S_{11}) and the comparison between the last block of 12-hour retention (S_{13}) and the first block of the 1-week retention test (S_{14}). Consolidation of sequence-specific was determined by comparing the sequence learning score of acquisition phase (R_9-Mean (S_8, S_{10})) with sequence learning score of 12-hour retention test (mean difference between R_{12} and the average of S_{11} and S_{13} (R_{12} – Mean (S_{11}, S_{13}))) and sequence learning score of 1-week retention test (mean difference between R_{15} and the average of S_{14} and S_{16} (R_{15} – Mean (S_{14}, S_{16}))) [13, 24, 37].

The acquisition data (General improvement) was subjected to a 2 (sleep, wake) × 2 (implicit, explicit) × block (R_1-S_{10}) Analysis of Variance (ANOVA) with repeated measures on block. Sequence-specific learning test was analyzed with a 2 (sleep, wake) × 2 (implicit, explicit) × block (R_9 and the average of S_8 and S_{10}) ANOVA with repeated measures on block. The 12 hour retention test (Consolidation of general learning) was analyzed with a 2 (sleep, wake) × 2 (implicit, explicit) × block 2 (S_{10}, S_{11}) ANOVA with repeated measures on block and the 7 day retention test was analyzed with a 2 (sleep, wake) × 2 (implicit, explicit) × block 2 (S_{13}, S_{14}) with repeated measures on block. Consolidation of sequence-specific learning was analyzed with a 2 (sleep, wake) × 2 (implicit, explicit) × block or sequence learning score (R_9-Mean (S_8, S_{10}), R_{12} – Mean (S_{11}, S_{13}) and R_{15}-Mean (S_{14}, S_{16})) ANOVA with repeated measures on block. Sidak post hoc test was used to conduct multiple comparisons of means in within-subjects effects in acquisition (General improvement of task) and sequence-specific consolidation phases. Also, in case of significant differences between groups (between-subjects effects) in consolidation of general sequence learning phase, Scheffe post hoc test was used. The df[2] were adjusted using the Greenhouse-Geisser correction in case of violation of the sphericity assumption, i.e. when the epsilon value was smaller than 1. Estimates of effect sizes were analyzed using partial eta squared (η_p^2). An alpha level of .05 was used for the analyses [24].

Results

Acquisition: General improvement of task

The analysis detected a main effect of block, for both element duration, $F_{5, 225} = 46.06$, $p < 0.01$, $\eta_p^2 = 0.51$, and error of prediction, $F_{5, 225} = 7.06$, $p < 0.01$, $\eta_p^2 = 0.21$. So that, they were higher in R_1, R_5, and R_9 than in the later Blocks ($S_{2–4}$, $S_{6–8}$, and S_{10}) where the repeated sequence was presented. The results indicated an improvement in speed and accuracy in repeated blocks (Figure and Table 1). Moreover, the Block × Awareness in error of prediction was also significant ($F_{6, 283} = 2.38$, $p < 0.05$, $\eta_p^2 = 0.05$). Figure and Table 1, show that explicit groups had lower error of prediction than implicit groups. All other main effects and interactions failed significance ($p \geq 0.05$).

Sequence-Specific learning

The analysis detected a main effect of block (R_9 – Mean (S_8, S_{10})), for both element duration, $F_{1, 44} = 147.52$, $p < 0.001$, $\eta_p^2 = 0.77$, and error of prediction, $F_{1, 44} = 5.78$, $p < 0.05$, $\eta_p^2 = 0.12$. So that, element duration and error of prediction in the R_9 (random sequence) were significantly higher than the average of S_8 and S_{10}. All other main effects and interactions were not Significant ($p \geq 0.05$, Figure 2).

Consolidation of general sequence learning

The results of the 2 × 2 × 2 ANOVA with repeated measures on block indicated a significant main effect of block for both element duration, $F_{1, 44} = 31.99$, $p < 0.01$, $\eta_p^2 = 0.42$, and error of prediction, $F_{1, 44} = 5.87$, $p < 0.05$, $\eta_p^2 = 0.12$. So that, the element durations and errors of prediction in S_{11} (beginning of session 2) were significantly

[2] Degrees of freedom.

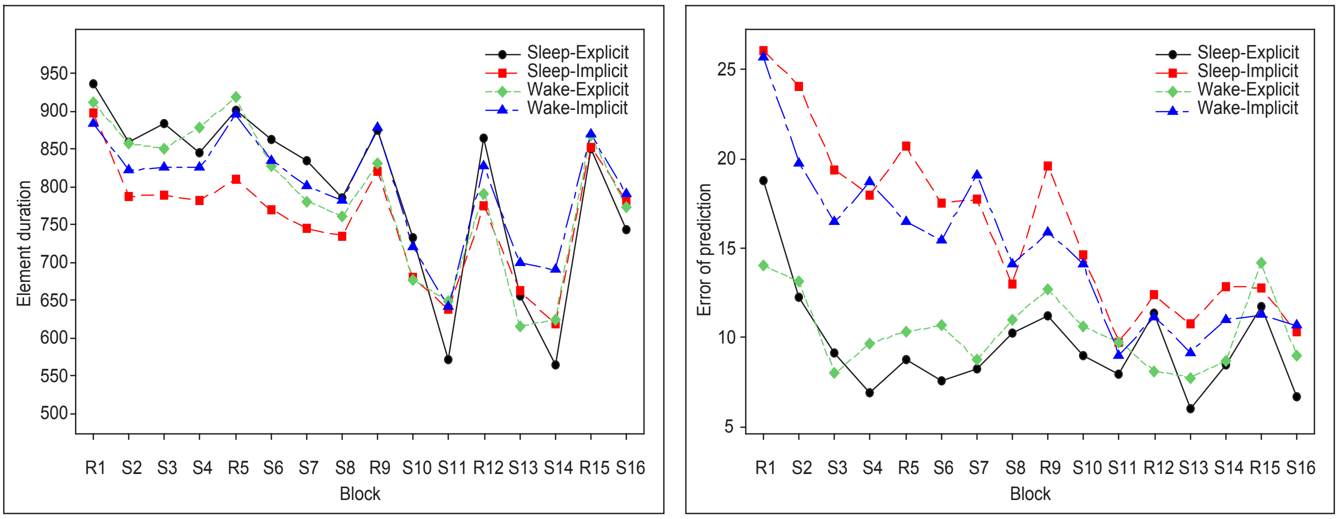

Figure 1. The mean of element durations and errors of prediction (mean of 96 trails) in 16 blocks (R_1 to S_{10} in acquisition phase, S_{11} to S_{13} after 12 hours and S_{14} to S_{16} after the 1-week test) in four groups. (R: Random block, S: Sequence block)

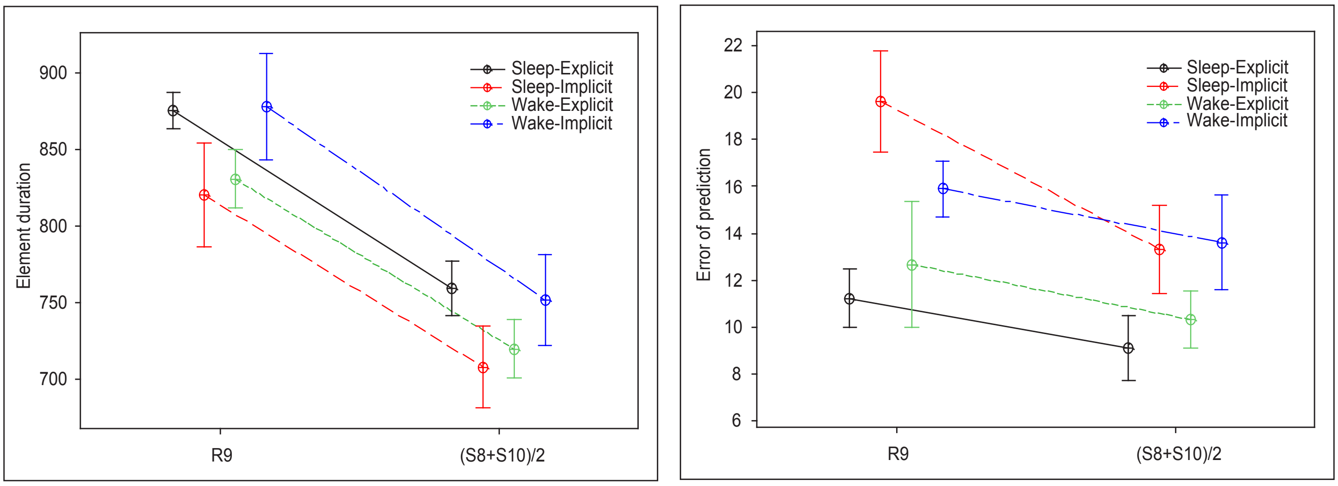

Figure 2. The mean and standard error of element durations (in msec) and errors of prediction between block 9 (R_9) and the average of blocks 8 and 10 (S_8, S_{10}) in acquisition phase for Sequence-Specific learning

lower than S_{10} (the last block of acquisition). Moreover, the Block × Sleep × Awareness in element durations were also significant ($F_{1, 44} = 9.59$, $p < 0.01$, $\eta_p^2 = 0.17$), in a way that, decreasing trend of sleep-explicit group was significantly different from that of sleep-implicit and wake – explicit groups (Figure 1). All other main effects and interactions failed significance ($p \geq 0.05$).

In addition, the results of the 2 × 2 × 2 ANOVA with repeated measures on block in 1-week retention test showed a significant effect of block for element duration, $F_{1, 44} = 7.75$, $p < 0.01$, $\eta_p^2 = 0.15$, indicating lower element durations at the beginning of session 3 (S_{14}) compared to the end of session 2 (S_{13}). The block × sleep was also significant, $F_{1, 44} = 7.42$, $p < 0.01$, $\eta_p^2 = 0.14$. So that, trend of element durations in sleep and wake groups were significantly different (Figure 1). Hence, the 1-week consolidation of general sequence learning was sleep-dependent. All main effects and interactions in error of prediction failed significance ($p \geq 0.05$).

Sequence-Specific Consolidation

The results of the 2 × 2 × 3 ANOVA with repeated measures on block (sequence learning score) indicated that the main effect of element duration was significant, $F_{2, 88} = 11.52$, $p < 0.01$, $\eta_p^2 = 0.21$. So that, the mean of sequence learning scores in session 2 (R_{12}-Mean (S_{11}, S_{13})) and session 3 (R_{15}-Mean (S_{14}, S_{16})) were significantly higher than sequence learning scores in acquisition phase (R_9-Mean (S_8, S_{10})). However, there was no main effect of block in error of prediction, $F_{2, 88} = 0.61$, $p \geq 0.05$. Hence,

Table 1. The mean and standard deviation of element duration and error of prediction (mean of 96 trails) in 16 blocks (R_1 to S_{10} in acquisition phase, S_{11} to S_{13} after 12 hours and S_{14} to S_{16} after the 1-week test) in four groups. (R: Random block, S: Sequence block)

Block		Element duration				Error of prediction			
		Sleep-Explicit	Sleep-Implicit	Wake-Explicit	Wake-Implicit	Sleep-Explicit	Sleep-Implicit	Wake-Explicit	Wake-Implicit
R_1	Mean	936.86	898.35	912.07	884.04	18.78	26.06	14.03	25.73
	SD	49.67	81.69	86.38	81.38	8.17	9.07	4.78	14.49
S_2	Mean	859.39	788.22	857.25	821.94	12.25	24.08	13.13	19.75
	SD	110.01	98.56	131.55	95.69	4.61	6.84	5.79	8.16
S_3	Mean	884.47	789.08	851.25	826.54	9.13	19.38	8.00	16.45
	SD	73.44	116.38	119.62	101.05	4.66	9.83	4.51	6.62
S_4	Mean	846.38	781.91	879.71	825.36	6.89	18.00	9.67	18.73
	SD	89.90	117.95	105.21	99.85	5.09	7.68	5.08	8.57
R_5	Mean	901.64	810.71	918.79	896.25	8.75	20.75	10.33	16.45
	SD	46.90	102.13	80.36	118.32	7.08	11.93	4.55	9.27
S_6	Mean	863.13	769.68	828.06	835.75	7.56	17.50	10.67	15.45
	SD	37.17	114.79	116.20	126.43	4.88	11.05	7.39	7.00
S_7	Mean	835.39	745.68	781.36	801.80	8.22	17.75	8.75	19.09
	SD	62.82	106.93	116.48	100.78	4.47	8.24	4.95	9.83
S_8	Mean	785.31	735.69	761.69	781.99	10.21	13.00	11.00	14.10
	SD	56.52	95.92	102.42	115.07	5.85	9.41	5.98	8.87
R_9	Mean	875.80	820.18	830.71	878.21	11.22	19.63	12.67	15.90
	SD	40.74	117.97	65.69	119.55	4.32	7.45	9.36	4.10
S_{10}	Mean	732.23	679.55	677.03	721.29	9.00	14.63	10.63	14.10
	SD	71.26	97.05	46.22	97.67	4.22	5.51	4.02	6.68
S_{11}	Mean	572.03	637.35	648.95	641.17	7.92	9.75	9.75	9.00
	SD	130.05	78.92	67.80	95.11	5.41	5.60	3.86	8.41
R_{12}	Mean	864.34	774.46	791.84	828.01	11.33	12.38	8.11	11.17
	SD	161.45	110.53	103.80	101.69	7.68	6.47	3.87	6.22
S_{13}	Mean	655.37	662.35	615.47	699.51	6.00	10.75	7.75	9.17
	SD	84.56	101.14	133.93	108.76	4.51	5.59	4.63	8.14
S_{14}	Mean	564.34	618.46	624.55	690.19	8.44	12.88	8.67	11.00
	SD	90.89	163.09	145.08	123.52	5.77	6.16	3.91	8.21
R_{15}	Mean	851.05	852.95	868.74	870.89	11.76	12.76	14.17	11.25
	SD	81.57	129.16	104.71	115.35	5.34	6.60	7.27	7.02
S_{16}	Mean	743.91	782.88	773.19	791.39	6.67	10.30	9.00	10.67
	SD	96.06	100.35	101.55	102.34	3.62	3.83	4.55	4.60

these results showed offline enhancement of sequence-specific learning after 12 hours only accrued in element duration.

The Block × Awareness in both element duration, $F_{2,\,88} = 4.07$, $p < 0.05$, $\eta_p^2 = 0.08$, and error of prediction, $F_{2,\,88} = 3.79$, $p < 0.05$, $\eta_p^2 = 0.08$, were significant. So that, the explicit groups recorded lower element durations of sequence learning scores in acquisition session (R_9-Mean (S_8, S_{10})) rather than the other conditions and they also showed higher errors of prediction of sequence learning scores in session 3 (R_{15}-Mean (S_{14}, S_{16})) rather than in acquisition session (R_9-Mean (S_8, S_{10}), Figure 3). Hence, these results indicated that the offline enhancement of sequence-specific learning was dependent on awareness. The block × sleep × awareness were also significant in element durations, $F_{2,\,88} = 3.41$, $p < 0.05$, $\eta_p^2 = 0.07$. In addition, it was the only sleep-explicit group that recorded higher element durations on sequence learning scores in

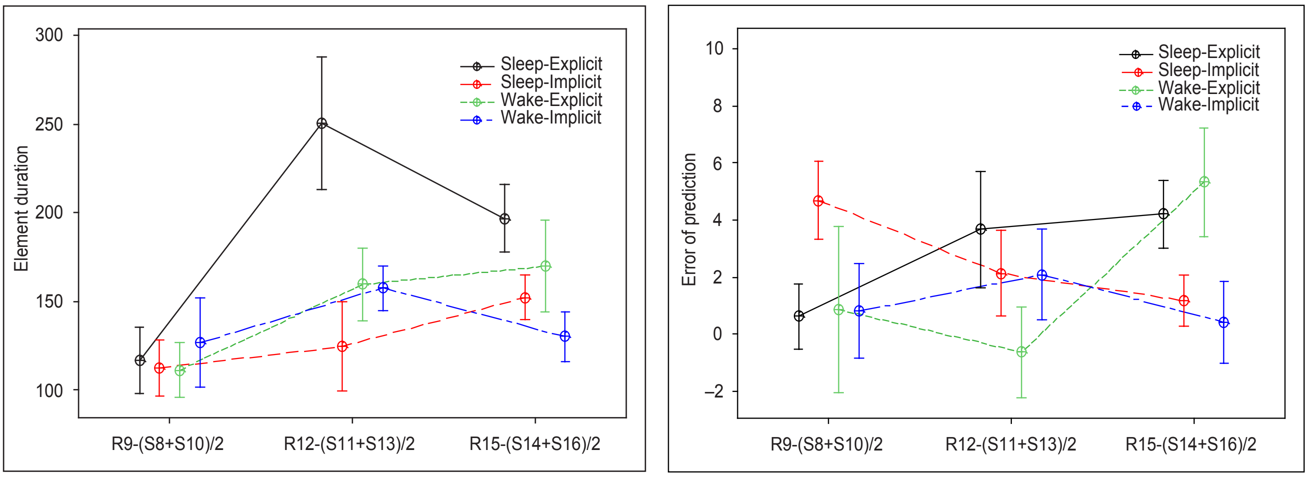

Figure 3. The comparison of mean and standard error of sequence learning scores (mean difference between Random block and average of the adjacent sequenced blocks) in acquisition phase (R_9-Mean (S_8, S_{10})), after 12hr (R_{12}-Mean (S_{11}, S_{13})) and after 1-week test (R_{15}-Mean (S_{14}, S_{16}))

session 3 (R_{15}-Mean (S_{14}, S_{16})) and session 2 (R_{12}-Mean (S_{11}, S_{13})) rather than in acquisition session (R_9-Mean (S_8, S_{10}), Figure 3). All other main effects and interactions were not Significant ($p \geq 0.05$).

Explicit knowledge

After completing the tests, four children from implicit groups correctly reported elements of the repeated sequence. Excluding these participants, the mean for the repeated sequence was 2.87 elements correct out of 12 which we take as chance level (expressing 4 elements or less than that was affected by the chance level) (SD = 0.93, and n = 20). Also, these children answered correctly in recognition test. As a result, they had explicit knowledge of sequence rules and therefore were omitted from the analysis.

Discussion

The purpose of this study was to investigate the role of sleep and awareness on motor memory consolidation with regard to general motor skill learning and sequence-specific learning by assessments of performance improvement between sessions in four groups (sleep-implicit, sleep-explicit, wake-implicit, and wake-explicit).

Performance on session 1 (Acquisition phase)

In the first session, children achieved general skill improvement in online learning by training the dynamic arm movement task in all the groups. The response time and error of prediction decreased in sequenced blocks. This session indicates that repetition and adequate practice in sequence skills increased speed and accuracy. These results implied that children's performance improves with more effort in training trails. So that, element duration and error of prediction in the acquisition phase from the first to last sequence blocks decreased [8, 24]. Wilhelm [41] showed that the average reaction times were decreased by training.

However, except awareness in the error of prediction, the type of knowledge and the training's start time in the acquisition phase (8 A.M. or 8 P.M) were not significant and general improvement occurred equally in all groups. Although explicit groups recorded higher element durations than implicit groups in acquisition phase, these differences were not significant. The current research was accordant with studies that had not observed any differences in response times between groups [5, 7, 21, 41]. This issue suggests that children can improve general motor performance both with and without receiving explicit instructions for the fixed sequence [37].

The improvement trend for the motor sequence in error of prediction (contrary to the speed) among implicit and explicit groups was different, such that the latter has a lower error. Despite this, with practice, the decreasing trend in the last sequence blocks was more than the first sequence blocks in implicit groups. This happened when the error in explicit groups during the practice had remained constant and low. Some researchers showed that being in the cognitive stage, could be the reason for the lower error of prediction in the explicit groups. The main characteristic of this stage was the conscious process of the information related to the task. They believed that explicit groups' awareness of the sequence pattern in error detection could be the reason they perform better than implicit groups. Due to implicit learning methods in implicit groups, they were not involved in cognitive processes. Therefore, their learning ability decreased in the performance error correction [21, 23].

Also, the results in the sequence-specific learning showed element duration and error of prediction in random block (R_9) were significantly higher than the average of the adjacent sequenced blocks (S_8 & S_{10}) in all of the groups. When the speed and error increased in unpredictable trials (random block) rather than predictable ones (repeated sequence blocks), sequence-specific learning occurred in children [24, 25]. In fact, when the random sequence was presented, the response time was increased and more errors occurred. This shows that children in implicit groups had learnt at least some of the sequence. Therefore, Knowing the rules and regularities of a task does not necessarily improve the performance and without that, the performance could be better than in random blocks [37].

Consolidation of general sequence learning

The results showed that children in all groups displayed slower response times and less errors of prediction after 12 hours. Only the block × sleep × awareness in elements duration were significant. So that, the decreasing trend of the elements duration in explicit-sleep group were significantly better than explicit-wake and implicit-sleep groups, and there were not any significant differences in other groups. When the sequence is learnt explicitly – rather than implicitly or time-based, the consolidation is sleep-dependent [2, 34]. The current results were consistent with some research in children, adolescence and adults which confirm that sleep had beneficial role in the process of declarative and cognitive procedural memory consolidation [3, 33, 35, 38, 41]. Sugawara et al. [35] expressed children's sleep is related to the improvement of motor sequence learning similar to adults. Wilhelm [41] and Peiffer et al. [31] also confirmed that sleep was the main factor of the declarative memory consolidation. Robertson et al. [33], Ashworth et al [3], and van den Berg et al. [38] mentioned that sleep would become the main role of the performance improvement, if participants had learned and practiced the regulation of the sequence task consciously.

Furthermore, when the children had practiced implicitly, the consolidation would be time-dependent. Some researchers, contrary to the recent findings, showed that sleep after training session could enhance the performance of motor skills [3, 11]. Cho et al. [11] revealed that adolescence's accuracy, similar to adults, enhanced after a night's sleep. Ashworth et al. [3] also confirmed that sleep would be beneficial in enhancement of children's procedural memory consolidation.

These contradictory results were probably related to the age, type, and nature of the task. Although, Ashworth et al. [3] used the procedural memory task for evaluating memory consolidation, only explicit aspects of the task are consolidated by sleep, not the implicit ones. Along this path, Janacsek and Nemeth [20] expressed that sleep-dependent procedural memory consolidation was task-related [20]. Moreover, participants of Cho et al. [11] research were adolescence, whereas in the current research, children were being studied. This is crucial, since age is an important factor in the sleep dependent memory consolidation of children [16].

These results were consistent with van den Berg et al. [38] which expressed that sleep has an efficient role in the cognitive process rather than a sequence task that is learnt implicitly. Wilhelm [41] also realized that children, unlike adults, showed less improvement in Finger Tapping Task after one night sleep in retention test compared to awakening. In addition, Fischer et al. [16] and Bothe et al. [7] showed the same results. Al-Sharman and Siengsukon [2] demonstrate that time, rather than sleep, appears to promote off-line learning of an implicit continuous motor task.

This evidence confirmed the active system consolidation theory [4, 6]. This theory expressed that during the early phases of procedural learning, memory is considered as an instable memory representation. Selected memory contents are reactivated during sleep and transferred into the long-term memory [4, 5]. Tononi and Cirelli [36] in Synaptic homeostasis hypothesis expressed that allowing the brain to go periodically "offline" must serve some important function. They suggested that during the subsequent sleep, when external inputs are reduced, slow oscillations renormalize these synapses inducing synaptic depression. This leads to a weakening of unimportant and less integrated information, making the important (signal) relative to the spurious information (noise) more salient. This process also restores the capacity of synapses to acquire new information. Robertson et al. [33] and Song [34] specified that offline improvement after initial training was affected by awareness. They mentioned that implicit and explicit offline learning are different and this difference was showed in the biological basis of acquisition of skills during training. These two types of learning are supported by two different mechanism: sleep-dependent and time-dependent mechanism. In this regard, Janacsek [19] expressed that cognitive functions were related to the frontal lobe. Normal sleep is related to the cognitive function. This showed that sleep could be effective on cognitive functions related to the frontal lobe and other areas of the cerebral cortex. Meanwhile, implicit learning related to the subcortical structure, had not benefit from sleep. Therefore, children in implicit groups activated subcortical structures. It is logical that their consolidation was time-dependent. Even though, the participants who learned the sequence consciously followed by sleep, showed improvement in the retention test [2, 34].

Furthermore, the results showed that consolidation of general sequence learning after 1-week occurred only in elements duration. Some studies on adults confirmed that consolidation of general learning was improved after 24-hours and one week [24, 25]. Desrochers et al. [13]

demonstrated that motor sequence learning benefited from sleep, but this was only evident after an extended period of time in children under six years old. Also the result showed that offline enhancement of general learning after 1-week was sleep-dependent. Element duration in sleep groups was decreased, but it was stable in wake groups. This issue showed that immediate sleep after acquisition of new skills would be an effective factor in persistence of information in the passage of time [13].

Consolidation of sequence-specific learning

These results showed that offline enhancement of sequence-specific learning was occurred only in the sleep-explicit group after 12 hours in element duration. However, the sequence-specific consolidation didn't occur in error of prediction. In this way, Nemeth et al. [26] and Meier and cock [24] showed that no improvement in sequence-specific learning was found in either age group, training session or time interval in implicit tasks.

Walker et al. [39] realized that sequence-specific consolidation occurred by using Finger Tapping Task which needed explicit knowledge of sequence. Van Abswoude et al. [37] reported that improvement was occurred only in accuracy (not reaction time) after 24 hours. Minimal differences were found between implicit and explicit condition. These contradictory results might be related to the age and task. Children in this study were under nine years old and the capacity of their working memory didn't effect in different types of learning. Children in the explicit group gained more sequence knowledge than the implicit group, and this knowledge did not transfer to a better sequence learning [37]. But in the current research, children in explicit group which experienced sleep immediately after training, gathered more information compared to implicit and wake groups in dynamic arm movement task. This matter caused a better transfer of sequence learning in 12 hours after the training. As a result, even if sleep didn't have an essential role in sequence-specific enhancement, it might be helpful in stabilization of memory traces besides other factors such as awareness.

Conclusion

The results of the current study showed that sleep wasn't the only essential factor to enhance offline motor sequence task for children after 12 hours, and their performance were related to both awareness and sleep. Offline enhancement of general sequence skill learning was sleep-dependent for explicit skills and time-dependent for implicit skills. Although sleep immediately after acquiring the new skills would be effective in information persistency in the passage of time. Concerning this issue, for future research, it is suggested to evaluate the other effective factors on a sleep-dependent general and sequence-specific consolidation of children such as nature and different types of tasks.

Conflict of interest: Authors state no conflict of interest.

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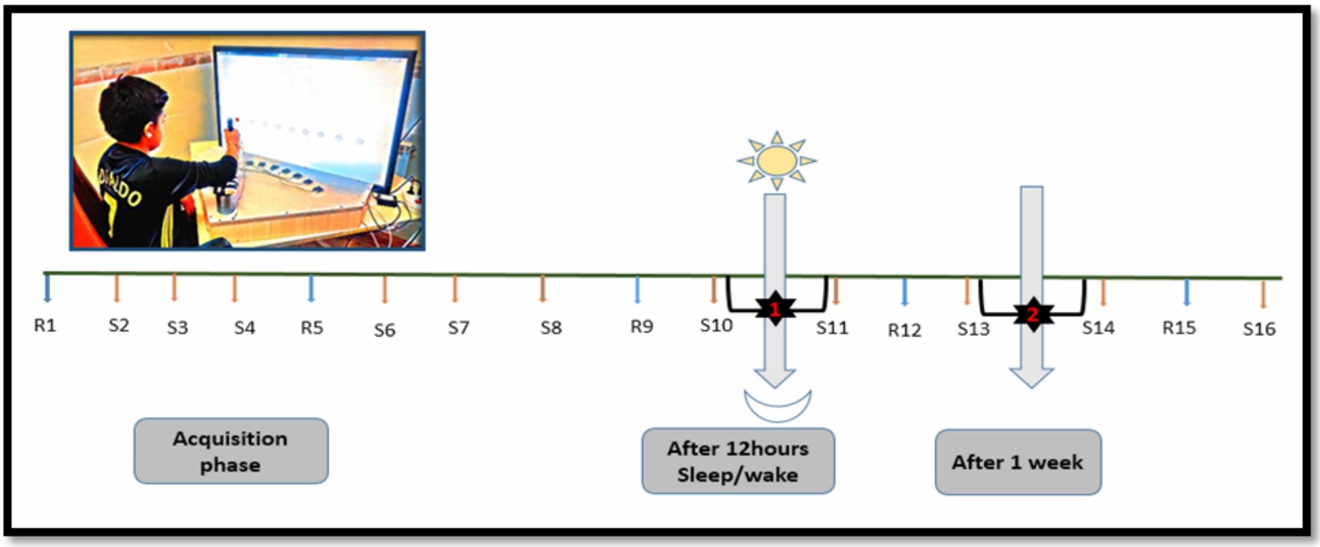

Appendix 1. Experimental design of sequence learning in three phases: acquisition phase (Blocks: R_1-S_{10}), 12-retention test (S_{11}-S_{13}) and 1-week retention or follow up test (S_{14}-S_{16}). Consolidation of general learning was determined by comparing S_{10} in acquisition phase and S_{11} in 12-hour retention test (1) and the comparison between S_{13} in 12-hour retention and S_{14} in 1-week retention test (2). Consolidation of sequence-specific learning was determined by comparing the sequence learning score of acquisition phase (mean difference between R_9 and the average of S_8 and S_{10}) with sequence learning score of 12-hour retention test (mean difference between R_{12} and the average of S_{11} and S_{13}) and sequence learning score of 1-week retention test (mean difference between R_{15} and the average of S_{14} and S_{16}).

Note: R: Random block, S: Sequence block.