Improved maze learning through early music exposure in rats

Frances H. Rauscher, K. Desix Robinson and Jason J. Jens

Department of Psychology, University of Wisconsin, Oshkosh, WI, USA

Rats were exposed in utero plus 60 days post-partum to either complex music (Mozart Sonata (K. 448)), minimalist music (a Philip Glass composition), white noise or silence, and were then tested for five days, three trials per day, in a multiple T-maze. By Day 3, the rats exposed to the Mozart work completed the maze more rapidly and with fewer errors than the rats assigned to the other groups. The difference increased in magnitude through Day 5. This suggests that repeated exposure to complex music induces improved spatial–temporal learning in rats, resembling results found in humans. Taken together with studies of enrichment-induced neural plasticity, these results suggest a similar neurophysiological mechanism for the effects of music on spatial learning in rats and humans. [Neurol Res 1998; **20**: 427–432]

INTRODUCTION

Several studies indicate that environmental enrichment can positively affect human cognition[1]. Specifically, musical experience may improve skills in related areas, particularly spatial domains. Musically trained children obtain higher scores on tasks requiring spatial abilities* than children provided with different training or no training[2–4]. These improvements are long-term, lasting at least one day. Similarly, subjects who listen to Mozart Sonata (K. 448) show significant short-term improvement (lasting 10–15 min) on spatial–temporal tasks[5–12] but not on spatial recognition tasks[5,13–16], when compared to control subjects who listen to something else or silence. Because spatial abilities have been shown to contribute to mathematical, scientific and artistic thought[17–19], this research has strong relevance for education and public policy.

Although the behavioral enhancement of spatial abilities following music exposure has been well established, little is known regarding the neural mechanism underlying the effect. Researchers have suggested that temporal-lobe structures are responsible for musical and spatial–perceptual representation[20,21] supporting the notion that these functions share partially overlapping neural substrates. Neural network models[22,23] in particular provide insight into the relationship. Specifically the 'trion' model[24], based on Mountcastle's[25,26] columnar organizational principles for mammalian cortex, proposes that musical activity strengthens inherent neural firing patterns that are also utilized by spatial–temporal tasks. Consistent with this model, EEG coherence data taken from subjects listening to either a Mozart Sonata (K. 448) or spoken text and then performing a spatial–temporal task revealed a carry-over of coherence patterns from the music (but not the text) to the spatial task in two cortical regions[27].

The search for a neural mechanism underlying the enhancement of spatial abilities following music exposure is complicated by the fact that it is neither desirable nor feasible to perform invasive studies exploring the anatomical effects of music on human brains. However, noninvasive studies using EEG, ERP, and brain imaging techniques, as well as studies of individuals who have suffered brain damage, have contributed to the understanding of the neural substrates of musical processing[28]. These experiments suggest that many brain areas are engaged during musical behaviors (some of these areas also serving nonmusical functions), with specific modules for musical abilities[29,30]. Further research indicates that music training affects the development of cortical areas. The left planum temporale is larger in musically trained than in untrained subjects[31], and the cortical representation of the fingers of the left hand is larger for string players than for controls[32]. Both effects were greater for those who began training at an early age. Although current imaging and EEG technology have revealed some of the areas of cortical activation that are associated with listening to music[27,29,33,34], researchers cannot determine if structural changes in the human brain can be induced by extensively listening to music without employing invasive techniques. The research reviewed above, however, strongly suggests a neural plasticity in humans that conforms to individual experience, and supports a neurophysiological basis for spatial task enhancement by music exposure[2–12].

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* Spatial ability is a construct containing several sub-classes, including the ability to manipulate objects in space and time, the ability to visualize varying configurations of a spatial array, the ability to determine spatial orientation with respect to one's body (as in maze running), etc.

Correspondence and reprint requests to: Frances H. Rauscher, PhD, Department of Psychology, University of Wisconsin, Oshkosh, WI 54901, USA. Accepted for publication March 1998.
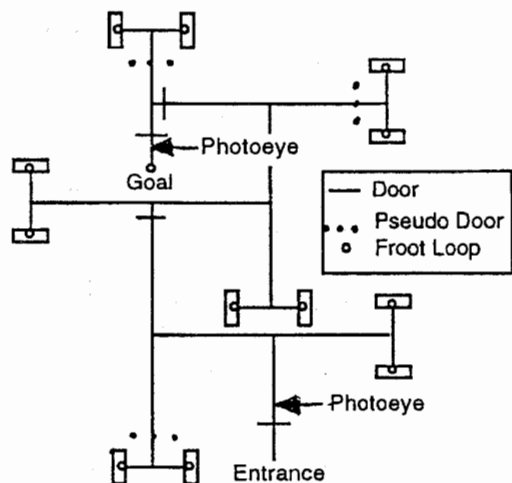
Figure 1: Floor plan of the T-maze, derived from a 12-unit maze developed by Stone and Nyswander[43]. The maze was constructed of $\frac{1}{4}$-inch gray plywood, with walls 15 cm high and alleys 15 cm wide. Pathways measured 50 cm. A Plexiglas top was fitted to the form of the alleys. Doors were suspended from strings attached to pulleys on the room's ceiling. Pseudo doors were included to make all pathways containing doors appear alike. Actual doors were closed behind the animal to prevent excessive retracing. A timer (Lafayette Instruments) was activated by two laser photoeyes attached to the maze wall 5 cm outside the start box (timer start) and 5 cm inside the goal box (timer stop). A Froot Loop™ (sugar cereal) was placed in the goal box and in vented enclosures at the end of each blind alley to prohibit the animal from scenting to the goal. The maze floor was washed with a solution of vinegar and water only as needed, permitting the rats to leave their odors in all parts of the maze. The room was lit by red light

Several researchers have explored the effects of enrichment on brain structure and function in rodents[35–37], demonstrating that enrichment-induced neurogenesis can improve maze performance[38]. Although the literature contains numerous studies exploring animal auditory processing, including pitch contour, timbre and rhythm[39–41] the neurobehavioral effects of auditory enrichment, particularly music, have received virtually no attention*.

To begin to identify the neural mechanism(s) governing music's enhancement of spatial reasoning in humans, we sought to establish an animal model for the effect employing *Rattus norvegicus* (the Long–Evans rat). Here we present data indicating that rats exposed to complex music performed better in a spatial maze than rats exposed to minimalist music, white noise, or silence.

MATERIALS AND METHODS
Animals were treated in accordance with the University of Wisconsin Oshkosh Institutional Animal Care and

*An exception is Schreckenberg and Bird[42], who found that the mouse hippocampus displayed abnormal neuronal overdevelopment as a function of exposure from birth to 60 days of rhythmically non-synchronized music. When tested in a maze, these mice performed worse than mice exposed to rhythmically synchronized music or silence.

Use Committee. We randomly assigned ninety rats *in utero* to three auditory exposure conditions (30 animals per group); (i) the *Allegro con spirito* of Mozart's *Sonata for Two Pianos in D Major*, (K. 448) (8 min 24 sec duration); (ii) the opening of Philip Glass' *Music With Changing Parts* (8 min 24 sec duration), or (iii) white noise. Breeder pairs were randomly placed in three separate exposure rooms (Mozart, Glass, white noise) prior to copulation. The male was removed three weeks after pairing, and ninety offspring (45 males and 45 females, 30 animals per exposure group) were randomly selected. Pups were housed post-weaning in same-sex pairs in 28 cm×21 cm×19 cm stainless steel cages. All animals were handled post-weaning for one minute daily to accustom them to human contact.

The music was looped through cassette players (Aiwa) to two speakers (Sony) placed on stools 122 cm from the animals' cages. White noise was produced by a white noise generator (Lafayette Instruments) and fed to a speaker (Realistic) mounted to the wall at ceiling level. Sound levels for all conditions registered 65–70 decibels at home cages, and were played during the animals' dark (active) cycles for 12 continuous hours per pay *in utero* (approximately three weeks), sixty days *post-partum*, and continuing through the last day of testing. Audio-cassettes were replaced as needed to maintain quality. In prior studies with humans, exposure to the Mozart work produced short-term enhancement of spatial–temporal reasoning when compared to the Glass composition (a particularly repetitive work) or silence[8]. White noise was chosen to mask extraneous sounds, and to provide the same magnitude of acoustic stimulation as the music conditions.

At age 58 days rats were weighed and put on a 23-hour food deprivation schedule to increase motivation, and were maintained at 85% free feeding weight. At 59 and 60 days animals were placed in the alley leading to the goal box of a six-unit T-maze (*Figure 1*) to familiarize them with the routine of maze running and food reward.

At 61 days we began testing maze performance. Ten rats from each Cage Exposure group were randomly assigned to one of three auditory Maze Exposure groups, Mozart, Glass or White Noise, yielding a three (Cage Exposure) by three (Maze Exposure) balanced design with ten animals per group. A trial ended after the rat reached the goal box or after four minutes, whichever came first. The sounds in the maze were produced by a Magnavox AC8848 cassette player with detachable speakers placed on opposite sides of the maze. Sound levels in the maze registered 65–70 decibels. Testing was performed blind over the course of five days, three trials per day, with ten-minute rest periods between trials. Home cage music exposure continued through the last day of testing. All trials were videotaped.

Dependent measures were total working time in the maze (WT) and total number of errors (E). WT was recorded electronically from timers wired to motion detecting laser photoeyes (see *Figure 1*), and errors were tabulated from videotapes by coders blind to experimental conditions. Two types of behavior were counted

as errors:

1. entrances of 10 cm or more into a blind alley, and
2. retracings of sections of the true path.

Inter-rater reliability was $r = 0.99$.

RESULTS

Three (Cage Exposure: Mozart, Glass, Noise) by three (Maze Exposure: Mozart, Glass, Noise) between-subjects analyses of variance (ANOVAs) performed separately on WT and E found main effects for Cage Exposure for both variables (*Table 1*). Unpaired two-tailed *t*-tests found that rats exposed to Mozart *in utero* and 60 days after birth reached the goal box more rapidly and made fewer errors than rats exposed to Glass or noise. Rats that received Glass and noise exposure did not differ from each other (*Figure 2*). No other effects or interactions were found, and Maze Exposure was therefore omitted from further analyses. *T*-tests revealed no sex effects for either variable.

We next examined WT and E by day. Two-factor (Cage Exposure, Day) mixed ANOVAs (with Cage Exposure a between-subjects factor and Day a within-subjects factor) revealed significant main effects for both factors (*Table 2*). No interactions were found. Individual one-factor (Cage Exposure) ANOVAs performed on each day revealed no differences in WT between groups on the first two days of testing (*Figure 3A*). On Day 3 the Mozart group differed significantly from the Glass group, a difference that increased through Day 5. The Mozart and Noise groups differed significantly on Days 4 and 5 only, whereas the Noise and Glass groups did not differ on any day. One-factor (Day) repeated measures ANOVAs for each exposure group indicated that, as expected, all groups showed significant learning during the course of the experiment.

Unlike WT, E for the Mozart group differed from the Noise group on Day 1, although the Mozart and Glass groups did not differ until Day 3 (*Figure 3B*). The Glass and Noise groups did not differ on any day. All

Table 1: Two-factor (Cage exposure, Maze exposure) Analysis of Variance for Working Time (WT) and Errors (E)

Source	df	F WT	F E
Cage exposure (C)	2	3.53*	10.42**
Maze exposure (M)	2	0.49	0.11
C×M	4	1.19	0.49
S within-group error	81	(513.21)	(1.36)

Values enclosed in parentheses represent mean square errors.
*S*, subjects; *, $p < 0.05$; **, $p < 0.01$.

Table 2: Two-factor (Cage exposure, Day) Analysis of Variance for Working Time (WT) and Errors (E)

	df	F WT	F E
Cage exposure (C)	2	9.07**	20.87**
Day (D)	4	24.48**	27.97**
C×D	8	1.06	0.81
S within-group error	1068	(1276.42)	(5.68)

Values enclosed in parentheses represent mean square errors.
S, subjects; *, $p < 0.05$; **, $p < 0.01$.
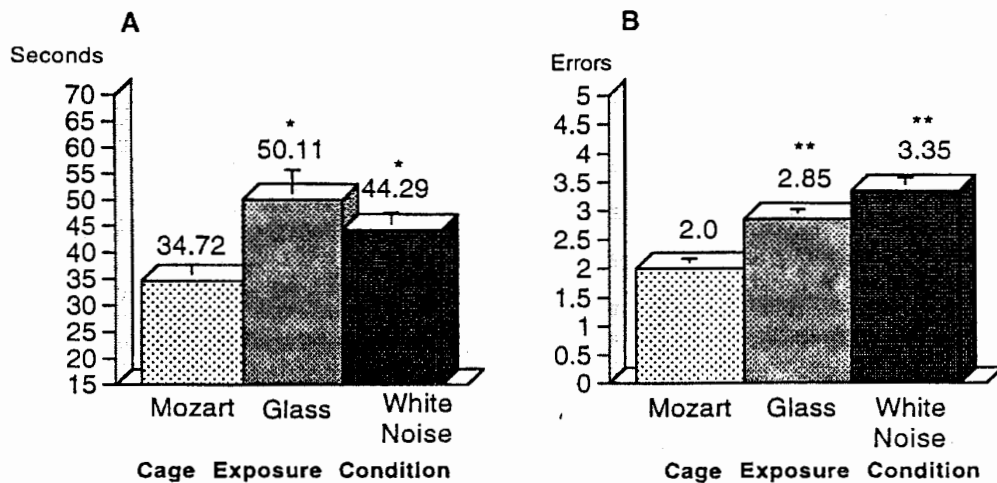
Figure 2: WT and E means and standard errors for rats who received cage exposure (*in utero* plus 60-days) to either Mozart, Glass or white noise. **A:** Animals in the Mozart group reached the goal box more rapidly than animals in the Glass ($t(1, 58) = 2.41$, $p < 0.05$) or Noise ($t(1, 58) = 2.13$, $p < 0.05$) groups. **B:** The Mozart group made fewer errors than the Glass ($t(1, 58) = 3.09$, $p < 0.01$) or Noise ($t(1, 58) = 4.78$, $p < 0.01$) groups. The Glass and Noise groups did not differ for either variable. *, $p < 0.05$; **, $p < 0.01$

A

B
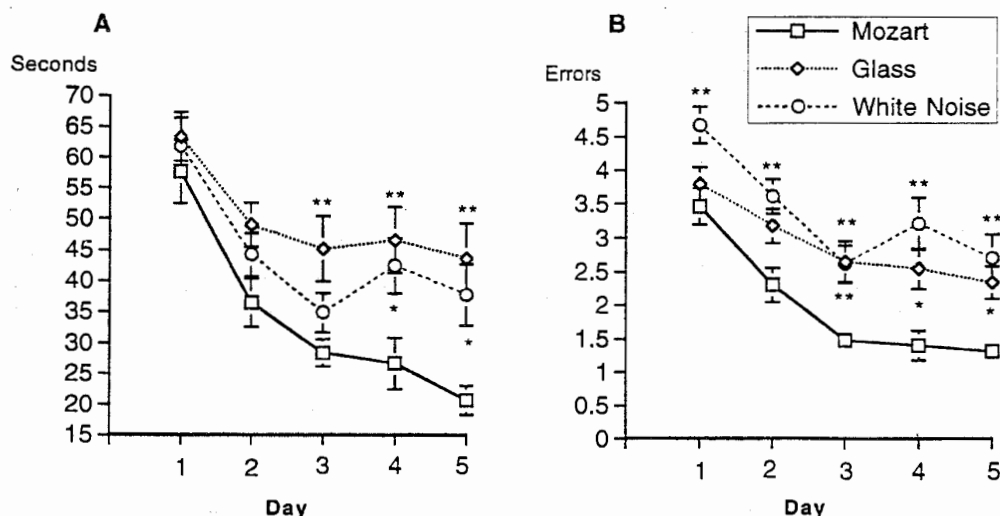
Figure 3: WT and E means and standard errors for each day of testing for rats who received cage exposure to Mozart, Glass or white noise. A: ANOVAs performed on each day revealed that the groups did not differ for WT on Day 1; however, the ANOVA began revealing significant differences on Day 3 $(F(2.267) = 5.07, p < 0.01)$, with the Mozart group differing from the Glass group $(t(2.178) = 4.99. p < 0.01)$. The Noise group began to differ from the Mozart group from Day 4 $(t(2,178) = 2.85, p < 0.05)$. The Glass and Noise groups did not differ on any day. B: For E, all ANOVAs performed on Day were significant. The Noise group made more errors than the Mozart group from Day 1 $(t(2.178) = 5.25, p < 0.01)$. On Day 3 $(F(2,267) = 6.85. p < 0.01)$, the Mozart group differed from both the Noise $(t(2.178) = 4.99, p < 0.01)$ and the Glass $(t(2.178) = 5.29, p < 0.01)$ groups. Significant differences were found through Day 5. We found no differences between the Glass and Noise groups on any day. *, $p < 0.05$; **, $p < 0.01$

groups made significantly fewer errors as the study progressed.

DISCUSSION AND CONCLUSION
The significant differences in learning between animals exposed to complex music (Mozart) *versus* minimalist music (Glass) or noise suggest that music exposure initiated at an early age enhances spatial performance in rats similar to results found in humans[5–12]. Rats exposed *in utero* plus 60 days to the same Mozart sonata that induced enhanced spatial–temporal task performance in humans performed better on a spatial task than genetically identical strains of rats exposed to Philip Glass' minimalist music or noise. The Mozart group completed the maze faster and made fewer errors overall. Although all groups significantly improved over days, the animals exposed to the Mozart sonata learned faster, as indicated by both WT and E.

It could be argued that the differences between the three groups were due to stress depressing the performance of the Glass and Noise groups rather than enrichment improving the performance of the Mozart group. To test this we ran additional subjects to compare the behavioral effects of Cage Exposure to white noise *versus* silence. Two groups ($n = 12$ and $n = 8$) were exposed to silence *in utero* and 60 days *post-partum*, and were then tested in the maze during exposure to either silence or white noise. A third group ($n = 8$) was similarly exposed to white noise and subsequently tested in silence. With the inclusion of the group previously

exposed to white noise in both cage and maze, this configuration yielded a two by two balanced design. A two (Cage Exposure: Noise, Silence) by two (Maze Exposure: Noise, Silence) ANOVA performed on WT and E revealed no differences between the Silence and Noise groups, suggesting that the performance of the animals exposed to Noise and Glass (which did not differ) was not induced by stress. Furthermore, one would expect stress-induced behaviors to manifest immediately, whereas WT for the three groups did not differ significantly until Day 3 (see *Figure 3A*). We found no body weight differences as a function of Cage Exposure, further reducing the likelihood that stress was a factor[44].

Although the dependent measures were correlated ($r = 0.75, p < 0.001$), it is possible that WT and E reflect different cognitive processes. For example, WT for the Mozart group did not differ significantly from the other groups until Day 3, suggesting that the effect of Mozart on WT reflected implicit perceptual learning. For E, the difference between the Mozart group and the Noise and Silence groups was evident on Day 1, perhaps suggesting a somewhat different process. Specification of the cognitive and neural mechanisms underlying this pattern of results is an enormous task that was not addressed by this experiment, and must await further research.

These data compliment findings demonstrating that environmental enrichment can induce hippocampal plasticity[35–37] and improve maze performance[38] in rodents. While it is tempting to conclude that the behavioral enhancement reported here was likewise

A

B

C

D
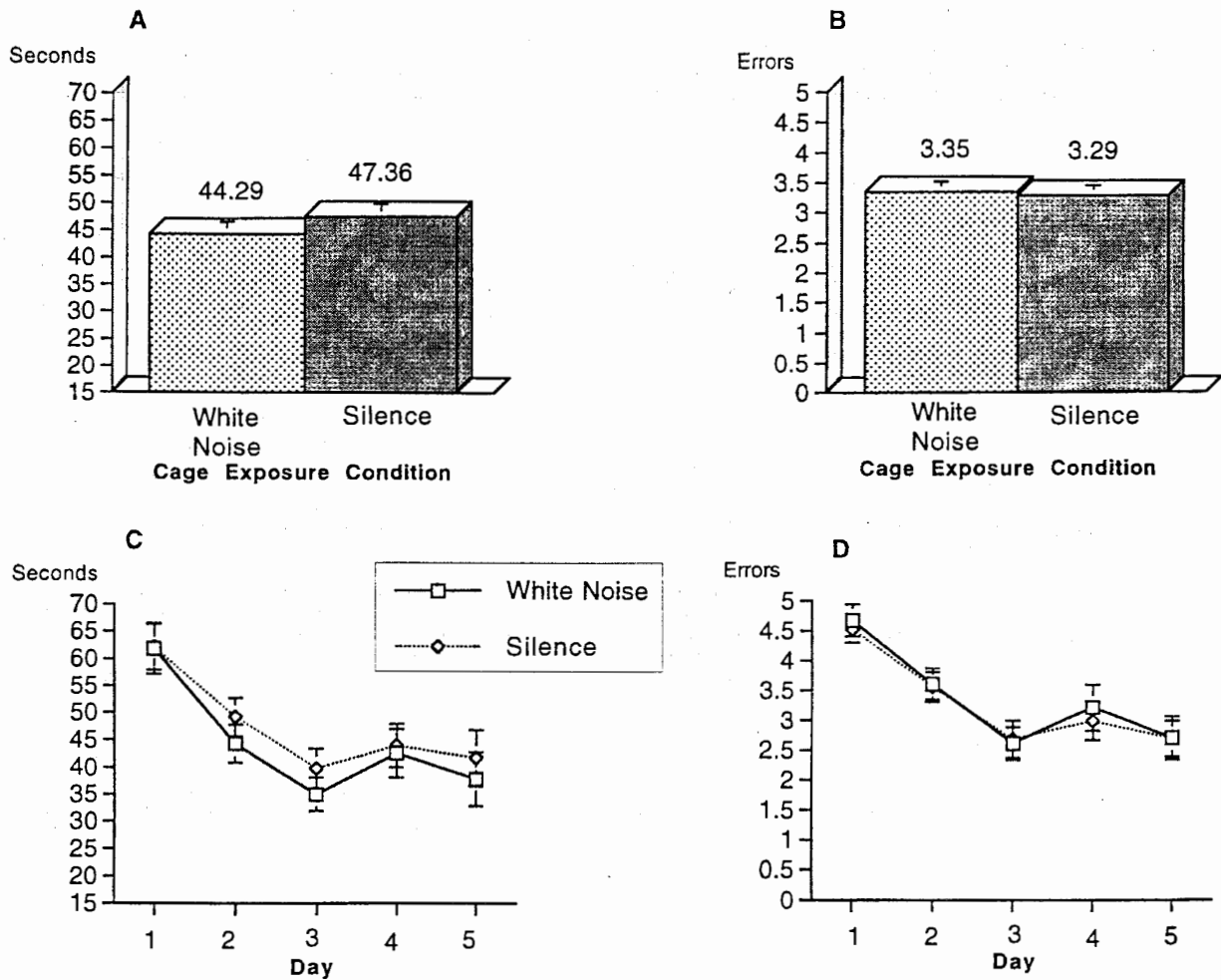
Figure 4: WT and E means and standard errors for the Noise and Silence Cage Exposure groups. **A: B:** Neither WT nor E differed for the two groups. **C: D:** Two-tailed *t*-tests performed on each day of testing found no differences between the Noise and Silence groups for either measure, with performance of the Silence group paralleling that of the Noise group. This reduces the likelihood that stress was a factor in the results and supports the Noise condition as a valid control

induced by hippocampal neurogenesis, we recommend a conservative approach. Further research must determine if and how the anatomy of the hippocampus was affected by musical enrichment, if other cortical areas were affected, and if nonspatial tasks were also influenced. Moreover, the physiological and behavioral effects produced by other forms of enrichment should be compared to those produced by music. It is also important to note, as pointed out by other researchers[37], that the 'enriched' environment in laboratory studies such as this must still be considered deprived compared to feral conditions. Therefore, questions of ecological validity should be addressed. Length and timing of exposure should also be explored. Finally, studies should determine the musical components that are relevant to the effect for both humans and rats.

By demonstrating that music induces improved spatial performance in the rat, this study lays the groundwork for further explorations into the neurophysiological factors that may mediate musical enhancement of spatial performance in humans. The work has strong implications for education and enrichment programs, and can potentially inform scientists about the relative contributions of biology and experience to human intelligence.

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