Effects of accelerated human music on learning and memory performance of rats

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Abstract

A total of forty, 7-week-old male rats were exposed to the ‘rodentized’ version (twice as fast as and one octave higher than the original) of Mozart’s Sonata for Two Pianos in D major for ten minutes a day for 10 weeks. One group (10 rats) received the musical stimuli before (‘B’), another during (‘D’) and the third before and during (‘BD’) the memory test, while the ten control (‘C’) animals were kept in silence. The animals’ spatial learning and memory ability was tested in an 8-arm radial maze. Rats exposed to the music showed a significant (7.1%) improvement in task acquisition (Group BD), but it did not practically change in Group D and worsened by 10.5% in Group B. The 2-h working memory significantly improved by 12.1% (BD) while practically did not change in Groups B and D. The reference memory improved by 11.9% in Group BD, but did not change in Group B and D, compared to the Control. The performance of the groups during the 4-h working memory test did not differ significantly. During the long-term test period the spatial memory performance of the music-exposed rats did not show significant differences compared to the Control (Table 1). At the same time, most results obtained in the long-term period were better than the corresponding short-term data. In conclusion, this particular piece of music, falling within the rats’ hearing range, was suitable for improving hippocampus-dependent spatial learning capacity, but only if the animals were exposed to it not only before but also during the task.

1. Introduction

The ‘Mozart effect’ (Rauscher et al., 1993) refers to a debatable (Pietzchnig et al., 2010) scientific theory, according to which listening to classical music may improve the subsequent learning ability and IQ test results of spatial-temporal character. However, the above meta-analysis did not care of the new neurophysiological and gene expression studies. Mozart’s Sonata for Two Pianos in D major, K 448 (hereinafter: the Sonata) and the Piano Concerto No. 23 (K 488) have been found to produce the most effective results. After listening to Mozart’s music, the mental functions of Alzheimer’s disease patients improved, and they were able to recall tunes (Fukui et al., 2012). The maze test performance of rats also improved under the influence of the Sonata (Rauscher et al., 1998). In contrast, silence, white noise or minimalist music have no similar effects. The question arises what type of music is generally able to produce the Mozart effect. Melodies similar in structure, harmony and predictability (recurrence) were also found to be efficient in producing similar effects. Hughes and Fino (2000) carried out computerized analysis of several pieces of music. In the case of Mozart and Bach, the long-term periodicity (the number of repetitions within 10–60 s, half a minute on average) is of decisive importance. The other common feature of Mozart’s and Bach’s music was their more frequent use of G3 (196 Hz), C5 (523 Hz) and H5 (987 Hz) sounds. During and shortly after listening to the Sonata the firing patterns of the neurons in the left temporal and right frontal cortex were synchronized (Rideout and Laubach, 1996). The effective musical stimuli induced more beta waves in the brain (Sarnthein et al., 1997).

Xu et al. (2009) kept newborn rats exposed to the Sonata eight hours a day for two months. According to their results, the ability to recognize sounds and the length of the sounds improved compared to the control group kept in silence. This ability was accompanied by increased expression of the NR2B subunit of the N-methyl-o-aspartate (NMDA) receptor. The NMDA receptors promote the maturation of glutamatergic synapses and ensure their plasticity during ontogenesis and learning.
Olton and Samuelson (1976) developed a maze of radial arms to measure the spatial learning and memory of rats. Since then the maze test has become a generally accepted method for studying the spatial memory of animals. According to Crusio and Schweger (2013), the maze test results are in strong correlation with the development of the hippocampal mossy fibre projections. In the field of spatial learning, there are two important types of memory, working memory and reference memory. While short-term memory means the storage of information for seconds, minutes, or a few hours, working memory refers to the short-time storage and manipulation of information for processing. In a maze test, working memory is responsible to remember the arms that have already been visited in search for food in the current trial. “Reference memory represents knowledge for aspects of a task that remain constant between trials.” (Nadel and Hardt, 2011). In a maze test, it means the knowledge about which arms of the maze always contain a food reward in each trial. In our recent work (Fekete et al., 2013), open-field (OF) activity and maze test performance were tested in rats after exposure to the Sonata. This particular music in the human hearing range was appropriate for improving hippocampus-dependent spatial learning capacity: it resulted in a significant improvement of the reference memory and memory return after the resting period, as well as a 4-h-long working memory retention compared to the group without music exposure. The main critical argument raised by Steele (2006) is that the hearing range of rodents is different from that of humans. The human audiogram extends from 0.02 up to 20 kHz, while that of rats from 0.5 up to approximately 90 kHz measured at 60–70 dB sound pressure level. He claims that rats cannot hear the lower 33–57% of the notes in this Sonata. Snowdon et al. (2015) emphasized, that music, used for animals, should be species-specific. In the experiments of Fekete and Bernitsa (2013) the ‘rodentized’ version of the Sonata (twice as fast as and one octave higher than the original) decreased kinetic activity and increased the time spent grooming and sitting more efficiently in an open-field trial than the original human version. The aim of the present work was to study the effect of ‘rodentized’ Mozart music as a typical classical piece on the spatial learning of rats, as well as on their memory retention performance in the 8-arm-maze. The present experimental question is the physiological-psychological effect on brain and not the possible subjective joy. Also, we wished to clarify how the timing of musical exposure (i.e. before, during, or before and during the memory test) might influence the results.

2. Material and methods

2.1. Animals, keeping, feeding, musical stimulus

After a five-day acclimatization (week 0) forty naïve male 7 weeks old SPF Crl:WI BR clinically healthy Wistar rats (breed by ToxiCoop Ltd., four groups, 10 rats/group) have been used. Three groups were exposed twice during the habituation period (for 80 min, once at 8 a.m. and once at 2 p.m.) daily to a 10-min-long arrangement of the Sonata in the rodentized version (one octave higher and twice faster, looped). For the treatment, the animals have been carried into a different room in individual carrying cages. Sample size was determined by Power Analysis, assuming a significance level of 5%, a power of 90% (Festing and Altman, 2002). The control (C) group (10 rats/group) was not exposed to any music, they spent the same time in a 3rd room, but in silence. From week 2, one group (10 rats) received the musical stimuli before the memory testing (before music = Group B), one group during the tasks (during music = Group D), and one group before and during the tests (before and during music = Group BD). The animals’ spatial learning and memory ability was tested in an 8-arm radial maze (Columbus Instruments, Ohio, USA). During the acclimatization periods, the rats were fed a standard (CRIL/N, Charles River Ltd) rodent diet of 11 MJ ME/kg energy density ad libitum, and they had continuous access to tap water. During the spatial learning and memory ability test, the daily ration was restricted (20–40 g/animal/day), in order to assure a live weight of 80–85% of the standard (Beatty and Rush, 1983). Food restriction is necessary in order to motivate the animals in this type of maze test. The rats were kept in groups of four, in polypropylene boxes (24 × 36 × 18 cm, with wire lids, AnimaLab) placed in a conventional animal room. There were 10 boxes, one animal from each group in every box. The rats were individually marked with animal marking spray (RAIDEX). As bedding wood chips have been used (Aspen wood bedding CLASSIC, AimaLab). The light-dark period was set to 12:12 h (dark phase from 6 a.m. to 6 p.m.), the applied light intensity was 60 lx, the room temperature 22 ± 3 °C, and the relative humidity 60 ± 5%. The music was of standard CD quality (44.1 KHz 16-bit resolution, ‘wav’ extension) with an attenuation of −6 dB SPL (sound pressure level). The one octave higher and two times faster version was produced by the NCH WavePad Audio Editing 2012 software; the change of overtones was neglected. During making the higher and faster (“rodentized”) version, the fine overtones remained the same.

2.2. Learning and behavioural studies

The testing (and the adaptation) was carried out in other separated rooms, with 40 dB background noise and 12–25 lx light intensity. Before the measurements, a 20-min adaptation time was applied in a resting room; the testing took place between 8:00 a.m. and 02:00 p.m in the dark phase. One box of rats has been carried at once into the resting room first. After the adaptation, one animal was carried into the test room, where the music-exposed group was exposed to the rodentized Sonata (10-min-long) before the test as a special acoustic stimulus of approximately 0.6–14 kHz pitch range. The SPL was 70 dB, using a Tamashi UX CD player and Genius speakers. Group D and BD were exposed to the same music during the test. The order of the animals from each box was the same: Group B, D, BD and C and the order of boxes was also constant during the different days and tests. The resting room and the testing room were far from each other, so animals in the resting room could not hear the music. After the test, the rat was put back into its homecage, so no animals were alone in the resting room, there was always at least three animals in a cage. Animals from group C also spent the same 10 min in the testing room before the maze test, but in silence. In the BD group, there was a pause in the music between the “before” and “during” phase while the animal was taken out from its cage and put into the maze.

The rats’ spatial learning and memory performance was studied using the maze test developed by Olton and Samuelson (1976) and modified by Vezer et al. (2000).

The labyrinth stands in the middle of a dimly lighted room without windows, its position is constant. During the test, some or all the arms is supplied by a bait (sweet popcorn) and rats are expected to remember the place of the bait, or to the place, where they found and ate it. Rats orientate and memorize the situation of arms and the fact of a visit in it using the breaking points of the maze, by the sight of the objects around the maze and by other extramaze cues, but smell. The bait cannot be seen from the centre. Odour-control orientation was distracted by cleaning the maze with disinfectant after every run.

Week 1: In the 1st week of treatment (Habituation), all animals had a 10-min training twice a day, adapting them to find food pellets in the maze arm ends. Perfect performance of this task required entering each arm only once. Week 2 (Acquisition of the task): During the 2nd week, the rats were first individually trained to learn the general cues of the task, that is, entering each one of the eight arms only once in a given session, with no more than one error per session, which was done with one training per day in six consecutive days. Acquisition errors consisted of revisiting an arm previously visited in the same session. This way, the rats were forced to learn a win-shift food search strategy. The percent rate of correct responses was counted as: (correct responses – acquisition errors) × 100, and was taken as performance indicator. All animals were expected to perform over 75%. Successful solution of the
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