

ORIGINAL ARTICLE

Childhood Music Training Induces Change in Micro and Macroscopic Brain Structure: Results from a Longitudinal Study

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Abstract

Several studies comparing adult musicians and nonmusicians have shown that music training is associated with structural brain differences. It is not been established, however, whether such differences result from pre-existing biological traits, lengthy musical training, or an interaction of the two factors, or if comparable changes can be found in children undergoing music training. As part of an ongoing longitudinal study, we investigated the effects of music training on the developmental trajectory of children's brain structure, over two years, beginning at age 6. We compared these children with children of the same socio-economic background but either involved in sports training or not involved in any systematic after school training. We established at the onset that there were no pre-existing structural differences among the groups. Two years later we observed that children in the music group showed (1) a different rate of cortical thickness maturation between the right and left posterior superior temporal gyrus, and (2) higher fractional anisotropy in the corpus callosum, specifically in the crossing pathways connecting superior frontal, sensory, and motor segments. We conclude that music training induces macro and microstructural brain changes in school-age children, and that those changes are not attributable to pre-existing biological traits.

Key words: auditory, child development, corpus callosum, cortical thickness, music training

Introduction

Playing music is a complex task that simultaneously involves motor, sensory, cognitive, and affective aspects. Of necessity, it requires the concurrent neural recruitment of distinct sensory systems, especially the auditory, somatosensory, and visual, as well as the interplay of these sensory systems with the motor, executive and affective systems. The mastering of this rich and

demanding process requires regular and intense practice, often from a young age, and the combination of such demands is likely to influence the differential development, maintenance, and operation, of certain brain structures and microstructures. Accordingly, over the past 2 decades, several investigators have reported differences in the brain and behavior of musicians compared to nonmusicians in adults and children (for

comprehensive reviews see [Gaser and Schlaug 2003](#); [Jäncke 2009](#); [Herholz and Zatorre 2012](#)).

Predictably, anatomical brain differences between musicians and nonmusicians have been found in auditory ([Schneider et al. 2002](#); [Gaser and Schlaug 2003](#); [Tillmann et al. 2003](#); [Luders et al. 2004](#); [Zatorre 2005](#); [Bangert and Schlaug 2006](#); [Jäncke 2009](#); [Herholz and Zatorre 2012](#); [Elmer et al. 2013](#); [Fauvel et al. 2014](#)), and sensorimotor areas ([Schlaug 2001](#); [Gaser and Schlaug 2003](#); [Jäncke 2009](#)). Of note, macro-structural differences have also been reported outside of auditory and sensorimotor-related regions including in inferior frontal regions ([Peretz et al. 2002](#); [Sluming et al. 2002](#); [Bermudez et al. 2009](#)), multimodal integration regions ([Münste et al. 2001](#); [Bangert and Schlaug 2006](#); [Zatorre et al. 2007](#)), and macro- and microstructural differences in the corpus callosum ([Schlaug et al. 1995b](#); [Schmithorst and Wilke 2002](#); [Bengtsson et al. 2005](#)), suggesting that music-related anatomical changes can extend to brain regions that are not primarily engaged by the immediate sensorimotor demands of music making.

Experience-dependent anatomical changes in musicians provide specific indication for the role of training. For example, [Bangert and Schlaug](#) reported that the amount of motor cortex devoted to the hand is larger in pianists than in nonmusicians, in both hemispheres, whereas in violinists the enlargement of the same region is seen primarily in the right hemisphere—the motor control region of the left hand ([Bangert and Schlaug 2006](#)). This fine anatomical difference is possibly related to the fact that pianists use both hands while playing (intense bimanual finger movements), whereas violinists primarily use their left hand to manipulate the strings and the right to control the bow (unilateral intense finger movement). Still, in the absence of longitudinal studies, the interpretation of such findings remains debatable. Specifically, the differences reported in cross-sectional studies, which mostly employ quasi-experimental designs, might be due to long-term regular and intensive training or might result primarily from pre-existing biological factors that would predispose an individual to develop musical aptitude if exposed to music during a sensitive period of development. Brain development is a dynamic process of change, shaped by complex genetic engagement and experience, both of which are at the mercy of environmental interventions, especially the latter ([Rosenzweig 2003](#)). The differences also might result from interaction or contributions of both training and pre-existing factors, with some differences relating more to training and others more primarily to pre-existing factors. One way of disentangling the effects of predisposing factors from those of musical training would be a longitudinal study of children beginning before the onset of their music training, and continuing on, into adulthood.

In spite of the desirability of longitudinal studies in this area, to date we are aware of only one such study, from [Schlaug and colleagues \(Schlaug et al. 2005\)](#). The scarcity is probably due to the difficulties, including recruitment and retention, facing researchers in this area. [Schlaug's](#) study showed no pre-existing anatomical brain differences in 6–7-year-old children who were about to begin musical training, and a matched group of children that were not intending to take music lessons. They reported that after 15 months of music training, children in the instrumental group showed structural changes, including increased gray matter density in the primary auditory and motor areas, and increased volume of the corpus callosum; not seen in the children who did not receive music training ([Hyde et al. 2009b](#)). However, no follow-up results have been reported. More recently, [Hudziak et al. \(2014\)](#) using longitudinal data from the

large NIH-MRI study of Normal Brain Development, showed that in healthy developing children playing a musical instrument was associated with more rapid changes of cortical thickness (CT) within areas implicated in motor planning and coordination; however, in the absence of pre-training data for the participants the possibility of pre-existing differences is a serious confound.

Taken together, the studies suggest that environmental training affects neural development. Nonetheless, the period of intervention, 15 months as reported by [Schlaug](#) appears to be rather short, and more uniform music training programs are essential to assess the strength and significance of changes related to music training on brain macro and microstructure during development. It is also important to further clarify whether changes observed in developing brains are limited to the territory directly involved in musical execution—sensory-motor and auditory areas—or if they encompass other territories. Lastly, including an active comparison group of children, engaged in a nonmusical but equally engaging and motivating activity can shed light on whether observed changes are specific to music or any other structured activity during development.

As part of an ongoing 5-year longitudinal study on the effects of music training on neural, cognitive and socio-emotional development in children from deprived socio-economic backgrounds ([Habibi et al. 2014](#)), we report both macro- and micro-structural brain changes in children after 2 years of musical training and compare them to 2 age-matched control groups, without music involvement but from the same socio-economic and cultural backgrounds ([Habibi et al. 2014](#)). The music training group was formed by children engaged in an El Sistema music education program of the Los Angeles Philharmonic. One control group involved children from the same socio-economic background and school system, who were participating in an athletic training program. This athletic training group was selected so as to control for those aspects of musical training that would be shared with a sport activity—a motivating, sustained, and engaging sensorimotor learning activity practiced in a group. The second control group involved children from the same socio-economic background and school system who were not part of any systematic or long-term music or sports training. They were not involved in any structured after school programs.

We used surface-based cortical reconstruction methods to examine the development trajectories of “cortical volume (CV)” and “cortical thickness” in selected regions of interest (ROIs). The ROIs were selected based on the previous literature to encompass regions that have been shown to have both near and far transfer effects of music training. We also used diffusion imaging to measure microstructural changes in white matter: fractional anisotropy (FA) and mean diffusivity (MD) of the cross-hemisphere connection, that is, in the corpus callosum and its subdivisions.

Materials and Methods

Subjects

Seventy-five children were recruited from public elementary schools and community music and sports programs within low-income communities of the greater Los Angeles area. Twenty children had enrolled and were about to begin their participation in the Youth Orchestra of Los Angeles at Heart of Los Angeles program (hereafter called “music group”). The

program is based on the Venezuelan system of musical training known as El Sistema and offers free music instruction 6–7 h weekly to children from underprivileged and low-income areas of Los Angeles. The program emphasizes ensemble practice and group performances, playing string instruments (violin and viola). Children applying to this program are selected, by lottery, up to a maximum of 20 per year, from a list of interested families. Nineteen children formed the first control group (hereafter called “sports group”) who had enrolled and were about to begin training in a community-based soccer program or a community-based swimming program and were not engaged in any musical training. The soccer program offers free soccer training 3 times a week with an additional game each weekend for children aged 6 and older; the swimming program offers free swim instruction 2 times a week to school-age children with an additional recreational swim session each weekend. Participants in both sports programs enrolled voluntarily in their respective programs and both programs were taught by trained coaches. Twenty-one children formed the second control group (hereafter called “control group”). Children in this second control group were recruited from public schools in the same Los Angeles area, provided they were not involved in any systematic and intense after-school program. Children in the music group were recruited first and we then recruited children volunteers from the sports and control group, via posted advertisement in the targeted communities, to match the number of recruited participants in the music group. Every child (family) that was interested was inducted provided they conformed to the basic induction criteria of age (6–7), basically healthy development, and not engaged in music activity. Recruitment was stopped as soon as the desired number of subjects was attained. All participants came from equally underprivileged backgrounds, with family incomes predominately below the Federal poverty guidelines and resided in geographical region of Los Angeles affected by extreme poverty levels, gang violence, and drug trafficking. Most child participants were of Latino background and were being raised in bilingual households. They attended English-speaking schools that did not offer comprehensive music or sports education programs. At all assessment times, participants were screened by interview with their parents to ensure that they did not have any diagnosis of developmental or neurological disorder and were tested with Wechsler Abbreviated Scale of Intelligence (WASI-II) (Wechsler 1999) to assure equal and normal cognitive development. Parents of children also answered an extensive structured interview on family income, education, and ethnicity, perceptions of child’s academic achievement and school participation, the child’s current and previous participation in extracurricular activities including involvement in sports or music programs, and the presence of any professional artists currently living in the child’s home. The structured interview was conducted in the parents’ preferred language, English, Spanish or Korean, by a research assistant who was a native speaker of that language (Habibi et al. 2014; Ilari et al. 2016).

Between the initial induction and 2 years later in the course of the longitudinal study, 15 participants (5 music, 4 sports-control, and 6 nonsports controls) discontinued their participation, in their respective program ($n = 4$) or the study ($n = 7$), relocated ($n = 2$), or no longer met safety criteria for MR imaging ($n = 2$) and thus were not included in the analysis. In addition, 4 participants (2 music, 1 sport, and 1 control) were excluded from the final analysis due to excess head motion during the MRI scan. Thus, here we report results from fifty-six participants. A power analysis with fixed effects model and alpha

level of 0.05, showed that the current sample size could yield a 70% probability to find a moderate effect size ($\eta^2 = 0.15$).

Recruitment and induction protocols were approved by the University of Southern California Institutional Review Board. Informed consent was obtained in writing, from the parents/guardians in the preferred language, on behalf of the child participants and verbal assent was obtained from all children individually. Either the guardians or the children could end their participation at any time. Participants (parents/guardians) received monetary compensation (\$15 per hour) for their child’s participation and children were awarded small prizes (e.g., toys or stickers).

Experimental Procedures

Handedness

Handedness was assessed as part of the Bruininks-Oseretsky Test of Motor Proficiency (BOT 2-Brief). Children were asked to write their name, throw a ball to the experimenter and kick a ball to the experimenter. Children were classified as right or left-handers if they used either left or right hand/foot for all three tasks. They were classified as mixed-handers if they used either left or right hand for only one of the tasks. In the music group, there was 1 left-handed boy; in the sports-control group, 1 left-handed girl; and in the nonsports control group, 2 left-handed boys.

Imaging

The brain imaging sessions included anatomical T1 (MPRAGE), diffusion, and functional MR imaging. Data collection and analysis for the MPRAGE and diffusion scans are described below; functional imaging results are reported separately (Sachs, Kaplan, Der Sarkissian and Habibi, under review). All MPRAGE images were handled according to the established policy of Dornsife Cognitive Neuroscience Imaging Center. For every child a T2 weighted scan is also obtained and is sent to a neuroradiologist for review. If an incidental finding is detected, the neuroradiologist contacts the physician designated specifically for that purpose by the family at the time they signed the informed consent and suggests further evaluation if needed. Two cases of incidental findings occurred during the study and corresponding participants’ data were excluded from analysis and are part of the list of dropouts mentioned earlier.

As described earlier (Habibi et al. 2014), we designed a child-friendly protocol that included a training session prior to the actual scanning session. Children learned about the scanner by watching a video and getting acquainted with a scanning session in a mock scanner while listening to the different types of sounds made by the scanner. During the actual scanning session, if children wished, one of the investigators remained in the scanner room and held the child’s hand. To assist children to remain motionless during the structural and diffusion scans, they watched a video of their choice. After the scanning session, children were shown and given an actual image of their brain.

High-resolution T1-weighted MPRAGE MRI images were acquired. There were 2 scanning sessions 2 years apart, referred to as “time 1” and “time 2”. At time 1, the scanner was a Siemens 3T Trio system equipped with a 12-channel head coil. We obtained an MPRAGE sequence with the following parameters: 1 mm × 1 mm × 1 mm resolution over a 256 mm × 256 mm × 208 mm FOV; TI/TE/TR = 800/3.09/2530 ms; flip angle = 10°; GRAPPA acceleration factor $R = 2$. Due to the difficulty of young children to remain still for extended periods of time, we

acquired two separate shorter MPAGE sequences (~3 min) instead of a single longer MPAGE acquisition we would typically use for adults. These 2 images were visually assessed for quality and motion artifacts, were registered and averaged. At time 2, the scanner was a Siemens 3T MAGNETOM Prisma System equipped with a 20-channel head coil; the following parameters were used: 1 mm × 1 mm × 1 mm resolution over a 256 mm × 256 mm × 256 mm FOV; TI/TE/TR = 850/32.05/2300 ms; flip angle = 8°; GRAPPA acceleration factor R = 2. For scan 2, we acquired one MPAGE sequence only as participants were able to remain still for the 5:32 min length of the scan.

Diffusion images were obtained during the same scan sessions as the high-resolution T1-weighted images, using the same scanners and coils for each time point as described previously. At time 1, we obtained 4 images without diffusion weighting ($b \approx 0$ s/mm²) and 80 diffusion weighted images (each with a b -value of 2500 s/mm², with orientations distributed as uniformly as possible based on the electrostatic repulsion approach described by Jones et al. 1999. These were acquired with a standard spin-echo EPI-based diffusion pulse sequence with the following parameters: transaxial slice orientation with A/P phase encoding, 2 mm × 2 mm in-plane resolution (128 × 128 matrix size) with 2 mm slice thicknesses, TE/TR = 115/10 000 ms, and 5/8th partial Fourier encoding with no parallel imaging acceleration. Instead of using the images produced by the scanner, we reconstructed images directly from the raw k-space data using SNR-enhancing joint reconstruction (Haldar et al. 2013), which has been demonstrated (both theoretically and empirically) to substantially reduce noise contamination while preserving resolution, experimental efficiency, and sensitivity to subtle microstructural changes (Haldar et al. 2013; Kim et al. 2016). Subsequently, images were corrected for geometric distortion using a separately-acquired B0 fieldmap and software from the BrainSuite Diffusion Pipeline (Bhushan et al. 2015; <http://brainsuite.org/processing/diffusion/>). At time 2, we obtained 7 images without diffusion weighting and the same 80 diffusion weightings from time 1. These were again acquired with a standard spin-echo EPI-based diffusion pulse sequence, but used the following parameters: transaxial slice orientation with interlaced A/P and R/L phase encoding (Bhushan et al. 2014) 2 mm × 2 mm in-plane resolution (128 × 128 matrix size) with 2 mm slice thicknesses, TE/TR = 85/10 000 ms, and 5/eighths partial Fourier encoding with no parallel imaging acceleration. As with time 1, these images were reconstructed directly from the raw k-space data using SNR-enhancing joint reconstruction (Haldar et al. 2013). Subsequently, distortion correction was performed by using a constrained reconstruction formulation (Bhushan et al. 2014) to simultaneously leverage the information available from multiple phase encoding directions. This was implemented using a customized version of the software available from <http://neuroimage.usc.edu/neuro/Resources/IPED>.

Analysis

Cortical Analysis

To analyze high-resolution T1 images, we used the BrainSuite software (Shattuck and Leahy 2002; <http://brainsuite.org/>) which incorporates a multiple step cortical surface extraction and labeling sequence for analyzing T1-weighted MR images. For scan 1, using a surface-constrained volumetric registration technique (SVReg, see Joshi et al. 2007, 2012), a customized child atlas was co-registered to each individual subject's brain. For each brain, transfer of region labels from the atlas yielded labeled cortical surfaces and volumes with segmentation of 31

individual cortical regions in each hemisphere. Each brain was examined after automated labeling and manual correction was applied whenever necessary due to edge mislabeling. BrainSuite's built-in "curve tool" was used to manually delineate corrected boundaries for all ROIs on the inner cortical surface. Macroscopic anatomical landmarks were used to visualize labels on the cortical surfaces and verified on the orthogonal volume planes. The processing sequence for scan 2 was similar with the exception that instead of using a customized common child atlas for registration purposes, we used each individual's manually label-corrected first scan as an atlas for the same subject in subsequent years. This method allowed improved registration of surfaces and labels.

We then determined gray matter volume and CT, for each hemisphere and each individual region of interest. These regions include: (1) anterior cingulate, (2) anterior dorsal cingulate, (3) posterior dorsal cingulate, (4) pars opercularis, (5) pars triangularis, (6) Heschl's gyrus (corresponding to primary auditory cortex), (7) anterior superior temporal gyrus, (8) posterior superior temporal gyrus (see Supplementary Method section 1 for details on the boundaries for delineating each ROI). Gray matter volume was computed using volumetric labels of the partial tissue fraction data generated by the BrainSuite and SVReg sequences which contains tissue fraction values corresponding to the percentages of gray matter, white matter and CSF in every voxel. Using these values, we computed the total amount of gray matter in each region of interest. Changes in CV for each specific region of interest was calculated by subtracting CV of the ROI at the time of the first scan from the CV of the ROI at the time of the second scan ($\Delta CV = CV1 - CV2$). We used a series of multivariate ANOVAs with music, control and sports as between factors to compare the changes in CV of the left versus the right hemisphere in the ROIs noted above.

CT was estimated using thickness PVC (Joshi et al. 2014), which incorporates tissue fraction and measured as the distance between inner cortical surface (the gray/white separation line), and pial surface. This measure was computed at every vertex on the surface mesh defining the 2 surfaces. The vertex-wise CT measure for all the vertices within a region of interest is averaged to produce the final CT value for that region of interest (Joshi et al. 2014). Cortical thinning in each specific region of interest was calculated by subtracting average CT of each ROI at the time of the scan 2 from the average CT of the ROI at the time of the scan 1 ($\Delta CT = CT1 - CT2$); As was done for CV, a series of multivariate ANOVAs with music, control and sports as between factors were used to compare the changes in CT of the left versus the right hemisphere in selected ROIs noted above.

Diffusion Analysis

We used the BrainSuite Diffusion Pipeline (<http://brainsuite.org/processing/diffusion/>) for analysis. For each subject, the diffusion MRI data was co-registered to the anatomical T1-weighted image using rigid registration based on the INVERSION method (Bhushan et al. 2015). Diffusion tensors were estimated using weighted linear least squares, and scalar diffusion parameters such as FA and MD were computed based on an eigendecomposition of the tensors as described by Kim et al. (2009). The FA and MD images subsequently were warped from the subject space to the common customized child atlas space using the one to one correspondence generated by SVReg between subject and the atlas. Mean and variance for FA and MD values were computed in the common space for

the whole corpus callosum (CC) and its seven subdivisions corresponding to the crossing of tracts from: (1) orbito-frontal and fronto-polar sectors (fronto.polar.orbito-CC); (2) superior frontal gyrus including the supplementary motor sector (SFG.SMA-CC); (3) precentral gyrus including the paracentral lobule (motor-CC); (4) post-central gyrus (sensory-CC); (5) temporal lobe (temporal-CC); (6) parietal lobe, mostly superior parietal lobule (SPL-CC); and (7) occipital lobe and precuneus (Occ.PreCun-CC) (see Supplementary Method section 2 for the boundaries for the Callosal subdivisions). Using a series of univariate ANOVAs with music, control and sports as between factors, we compared FA and MD for scans 1 and 2 separately, in the seven segments of the corpus callosum noted above.

Selection of ROIs

We hypothesized that brain changes in the a priori ROI within the auditory cortex, the inferior frontal and the corpus callosum would be influenced by music training. Auditory regions, including the primary and association auditory areas, are known to be of critical importance in processing of acoustic information, including spectral and temporal cues, in controlling attention-related auditory process (Jäncke et al. 2001) and providing continuous feedback for appropriate motor execution during musical performance, a key skill in music making (Zatorre and Belin 2001; Zatorre et al. 2002; Peretz and Zatorre 2005). Regions in the inferior frontal gyrus (pars opercularis and pars triangularis), have been shown to be involved during, audiospatial and visuospatial localization (Haxby et al. 1991; Martinkauppi et al. 2000), auditory-motor mapping (Lahav et al. 2007), musical syntax processing (Koelsch 2005), and sight reading (Sluming et al. 2002), all skills that are practiced and mastered during music learning, and which are essential for playing a musical instrument. Finally, playing a musical instrument also requires intense and independent bimanual motor training and control, which has been shown to have an impact on callosal microstructure and interhemispheric connectivity. Specifically, differences related to music training have been reported in the anterior segment of corpus callosum which contains fibers connecting the frontal cortices, including the premotor and supplementary motor areas (Schlaug et al. 1995b), and in the mid-body section of the callosum, comprising fibers that connect primary sensorimotor cortices (Hyde et al. 2009a). Because these specific predictions were a priori and were motivated by multiple previous reports in children and adult cohorts (Schneider et al. 2002; Schlaug et al. 2005; Hyde et al. 2009a; Bailey et al. 2014), we did not control for multiple comparisons for any of the statistical tests for ROI analysis. ROI within the cingulate gyrus (anterior cingulate, anterior dorsal cingulate, and posterior dorsal cingulate) were selected to provide a comparison between regions that are influenced directly (near transfer effect) or indirectly (far transfer effect) by music training.

MRI Scanner Hardware Upgrade

The MR scanner hardware upgrade (from a Siemens 3 T Trio to a Siemens 3 T Prisma) between the 2 scans was not expected to have a significant impact on the structural T1-weighted images. We assessed the impact of the system hardware upgrade (Siemens 3 T Trio to Siemens 3 T Magnetom Prisma) and change in acquisition parameters on cortical measures between two scans by acquiring MPAGE images in two healthy adult participants before and after the system upgrade

(1 month apart). In healthy adults, short-term changes in cortical gray matter are not typically expected. The results confirmed that the upgrade had minimal and nonsignificant impact on measures of CT and volume in these adult participants. Therefore we estimated longitudinal changes of CT and volume, for each ROI, by subtracting the average of each variable at scan 2 from scan 1.

However, the change in scanner hardware posed a bigger problem for diffusion sequences. Specifically, the diffusion encoding parameters that were used to achieve the b -value of 2500 s/mm^2 (the gradient amplitudes and timing parameters Δ and δ) were expected to change substantially between the 2 scans because of the difference in gradient performance between the Trio and Prisma. The diffusion signal is known to be time dependent and such changes in diffusion encoding would present a substantial confound for longitudinal analysis. Therefore, measures of FA and MD of the white matter for each ROI were estimated separately for each scan and no attempt was made to directly compare data from scans 2 to 1.

Results

The final number of participants included in the analysis was 18 children (5 girls and 13 boys, mean age at baseline assessment = 80 months, $SD = 6.64$) in the music group. Eighteen children (10 girls and 8 boys, mean age at baseline assessment = 83.2 months, $SD = 7.04$) in the sports-control group and twenty children (8 girls and 12 boys, mean age at baseline = 85.5 months, $SD = 5.82$) in the control group.

There was a significant difference in age among the three groups, at the time of scan 1F (2, 53) = 4.56, $P = 0.014$, $\eta^2 = 0.14$; where children in the music group were on average 6 months younger than the children in the control group (M vs. C, $P = 0.01$). The age difference between children in the music and sports group (M vs. S, $P = 0.2$) or children in the sports and control group (S vs. C, $P = 0.5$) was not significant. At the time of scan 2, children in the music group were still on average 4 months younger than the children in the control group although the main effect of age was no longer significant F (2, 53) = 1.60, $P = 0.1$, $\eta^2 = 0.08$, among the three groups. This was due to the interval between the two scans; on average, it was 1.8 months longer for the children in the music than in the control group [Music: 24.8 (0.78) months; Sports: 24 (1.02) months and Control: 23 (0.91) months]. There was no difference in sex distribution among the three groups χ^2 (2) = 1.91, $P = 0.38$. The average IQ (FSIQ-2) across all participants, at the time of Scan 1, was 98.1 (11.6), [Music: 101.7 (13.4); Sports: 96.9 (8.4) and Control: 95.9 (12.1)]. The average IQ (FSIQ-2) across all participants, at the time of Scan 2, was 97.6 (12.9), [Music: 101.8 (15.4); Sports: 95.9 (10.8) and Control: 95.1 (11.8)]. There was no difference in IQ scores (FSIQ-2), among the three groups neither at the time of scan 1, F (2, 53) = 1.35, $P = 0.26$, $\eta^2 = 0.04$, nor at scan 2, F(2, 52) = 1.46, $P = 0.24$, $\eta^2 = 0.05$. IQ, age, sex, or interval between the 2 scans were not included as factors in subsequent analysis.

Cortical Thickness

Hemispheres

At time 1, across all participants ($n = 56$) the average CT of the brain was 3.22 mm (min = 2.81 to max = 4.21) for the left hemisphere and 3.22 mm (min = 2.89 to max = 4.26) for the right. At time 2, it was 2.81 mm (min = 2.29 to max = 3.09) for the left hemisphere and 2.88 mm (min = 2.4 to max = 3.25) for the right.

From scans 1 to 2, cortical thinning was significant over both the left ($F(1, 53) = 111.75, P < 0.001, \eta^2 = 0.67$) and right ($F(1, 53) = 78.128, P < 0.001, \eta^2 = 0.59$) hemispheres. The group \times year interaction, however, was not significant for either left ($F(2, 53) = 1.42, P = 0.25, \eta^2 = 0.05$) or right hemispheres as a whole ($F(2, 53) = 1.83, P = 0.16, \eta^2 = 0.05$). All 3 groups appeared to have a similar degree of thinning at hemisphere level; the average cortical thinning for the left hemisphere was 0.4 mm (music group, $n = 18$), 0.5 mm (sports group, $n = 18$) and 0.3 mm (control group, $n = 20$); for the right hemisphere it was 0.3 mm (music), 0.4 mm (sports), and 0.26 mm (control).

ROI Analysis

1. Anterior Cingulate: there was no significant difference between the left and right anterior cingulate $F(1, 53) = 0.39, P = 0.53, \eta^2 = 0.007$, nor between groups $F(2, 53) = 2.56, P = 0.08, \eta^2 = 0.08$. However, there was a significant interaction of group \times laterality $F(2, 53) = 4.32, P = 0.018, \eta^2 = 0.14$, driven by the larger reduction of CT in the left anterior cingulate in the sports compared to the music and control groups; however, none of the post hoc comparisons remained significant nor marginally significant indicating that the driver of this effect was somewhat increased rate of thinning of left versus right anterior cingulate in the sports group with the opposite direction in the control group.
2. Anterior dorsal cingulate: there was no significant difference of group $F(2, 53) = 1.53, P = 0.22, \eta^2 = 0.05$, laterality $F(1, 53) = 2.66, P = 0.1, \eta^2 = 0.04$ or group \times laterality interaction $F(2, 53) = 0.37, P = 0.69, \eta^2 = 0.01$.
3. Posterior dorsal cingulate: there was no significant difference of group $F(2, 53) = 0.20, P = 0.81, \eta^2 = 0.007$, laterality $F(1, 53) = 0.63, P = 0.43, \eta^2 = 0.01$, or group \times laterality interaction $F(2, 53) = 0.70, P = 0.49, \eta^2 = 0.02$.
4. Pars opercularis: there was no significant group difference $F(2, 53) = 1.29, P = 0.28, \eta^2 = 0.04$. Of note, the cortical thinning of the left pars opercularis was significantly larger than that in the right $F(1, 53) = 21.5, P = 0.000, \eta^2 = 0.28$, however, the group \times laterality interaction was not significant $F(2, 53) = 0.33, P = 0.71, \eta^2 = 0.01$.
5. Pars triangularis: there was no significant group difference $F(2, 53) = 0.46, P = 0.63, \eta^2 = 0.01$; as was the case for the pars opercularis the cortical thinning on the left was significantly larger than that of the right $F(1, 53) = 7.49, P = 0.008, \eta^2 = 0.12$, but the group \times laterality interaction was again not significant $F(2, 53) = 0.50, P = 0.60, \eta^2 = 0.01$.
6. Heschl's gyrus: there was no significant group difference $F(2, 53) = 0.65, P = 0.52, \eta^2 = 0.02$; as described for the 2 prior cases, the cortical thinning on the left was significantly larger than on the right $F(1, 53) = 10.13, P = 0.002, \eta^2 = 0.16$, however, the group \times laterality interaction was not significant $F(2, 53) = 1.44, P = 0.24, \eta^2 = 0.05$.
7. Anterior superior temporal gyrus: there was no significant group difference $F(2, 53) = 0.21, P = 0.80, \eta^2 = 0.007$; once again, the cortical thinning on the left was significantly larger than on the right $F(1, 53) = 6.02, P = 0.01, \eta^2 = 0.1$, but the group \times laterality interaction was not significant $F(2, 53) = 0.29, P = 0.74, \eta^2 = 0.01$.
8. Posterior superior temporal gyrus: there was no significant difference of group $F(2, 53) = 0.14, P = 0.86, \eta^2 = 0.0025$ or laterality, $F(1, 53) = 1.11, P = 0.29, \eta^2 = 0.02$; However, here there was a significant group \times laterality interaction $F(2, 53) = 3.99, P = 0.024, \eta^2 = 0.13$, (see Fig. 1); while the difference of

cortical thinning between left and right posterior superior temporal gyrus was not different for the 2 control groups (sports and control), the music group showed a trend larger thinning of the left versus right posterior superior temporal gyri (Music: Left vs. Right $P = 0.14$; Sports: Left vs. Right, $P = 0.98$ and Control, Left vs. Right $P = 0.69$). Other post hoc comparisons were not significant.

Cortical Volume

Hemispheres

At time 1, across all participants ($n = 56$) the average CV of the left hemisphere was 309.24 cc (min = 251.73 cc to max = 370.38 cc) and 311.63 cc (min = 257 cc to max = 370 cc) for the right hemisphere. At time 2 it was 290.09 cc (min = 229.41 cc to max = 347.91 cc) in the left hemisphere and 298.18 cc (min = 240 cc to max = 357.8 cc) for the right. Longitudinal assessment of change in gray matter volume revealed significant loss of CV over the left hemisphere ($F(1, 53) = 57.72, P < 0.000, \eta^2 = 0.52$) and the right hemisphere ($F(1, 53) = 33.22, P < 0.000, \eta^2 = 0.38$) from scans 1 to 2. The group \times year interaction, however, was not significant for left hemisphere ($F(2, 53) = 0.92, P = 0.40, \eta^2 = 0.03$), or right hemisphere ($F(2, 53) = 0.80, P = 0.45, \eta^2 = 0.02$). All groups had a similar degree of CV decrease at hemisphere level; the average volume decrease was 20.15 cc (music group, $n = 18$), 23 cc (sports group, $n = 18$) and 14.76 cc (control group, $n = 20$); for the right hemisphere it was 13.3 (music), 17.28 cc (sports), and 10.03 cc (control).

ROI Analysis

1. Anterior Cingulate: there was no significant group difference $F(2, 53) = 1.34, P = 0.26, \eta^2 = 0.04$, laterality $F(1, 53) = 0.22, P = 0.63, \eta^2 = 0.004$ or group \times laterality interaction $F(2, 53) = 1.02, P = 0.36, \eta^2 = 0.03$.
2. Anterior dorsal cingulate: there was no significant group difference $F(2, 53) = 2.08, P = 0.13, \eta^2 = 0.07$, laterality $F(1, 53) = 2.34, P = 0.13, \eta^2 = 0.04$ or group \times laterality interaction $F(2, 53) = 0.12, P = 0.88, \eta^2 = 0.004$.
3. Posterior dorsal cingulate: there was no significant group difference $F(2, 53) = 0.24, P = 0.78, \eta^2 = 0.009$, or group \times laterality interaction $F(2, 53) = 1.24, P = 0.29, \eta^2 = 0.04$. However, CV decrease was significantly larger on the right posterior dorsal cingulate compared to the left $F(1, 53) = 0.1787, P = 0.00, \eta^2 = 0.25$.
4. Pars opercularis: there was no significant group difference $F(2, 53) = 1.08, P = 0.34, \eta^2 = 0.03$; although the decrease in CV in the left pars opercularis was significantly larger than in the right $F(1, 53) = 6.67, P = 0.01, \eta^2 = 0.11$, the group \times laterality interaction was not significant $F(2, 53) = 0.82, P = 0.44, \eta^2 = 0.03$.
5. Pars triangularis: there was no significant group difference $F(2, 53) = 0.29, P = 0.74, \eta^2 = 0.01$, laterality $F(1, 53) = 0.69, P = 0.40, \eta^2 = 0.01$ or group \times laterality interaction $F(2, 53) = 1.03, P = 0.36, \eta^2 = 0.03$.
6. Heschl's gyrus (Transverse Temporal Gyrus): there was no significant group difference $F(2, 53) = 1.58, P = 0.21, \eta^2 = 0.05$; CV decrease on the left was significantly larger than on the right $F(1, 53) = 10.696, P = 0.001, \eta^2 = 0.16$, however, the group \times laterality interaction was nonsignificant $F(2, 53) = 0.92, P = 0.40, \eta^2 = 0.03$.
7. Anterior superior temporal gyrus: there was no significant group difference $F(2, 53) = 0.20, P = 0.81, \eta^2 = 0.007$, laterality $F(1, 53) = 0.36, P = 0.54, \eta^2 = 0.006$ or group \times laterality interaction $F(2, 53) = 1.93, P = 0.15, \eta^2 = 0.06$.

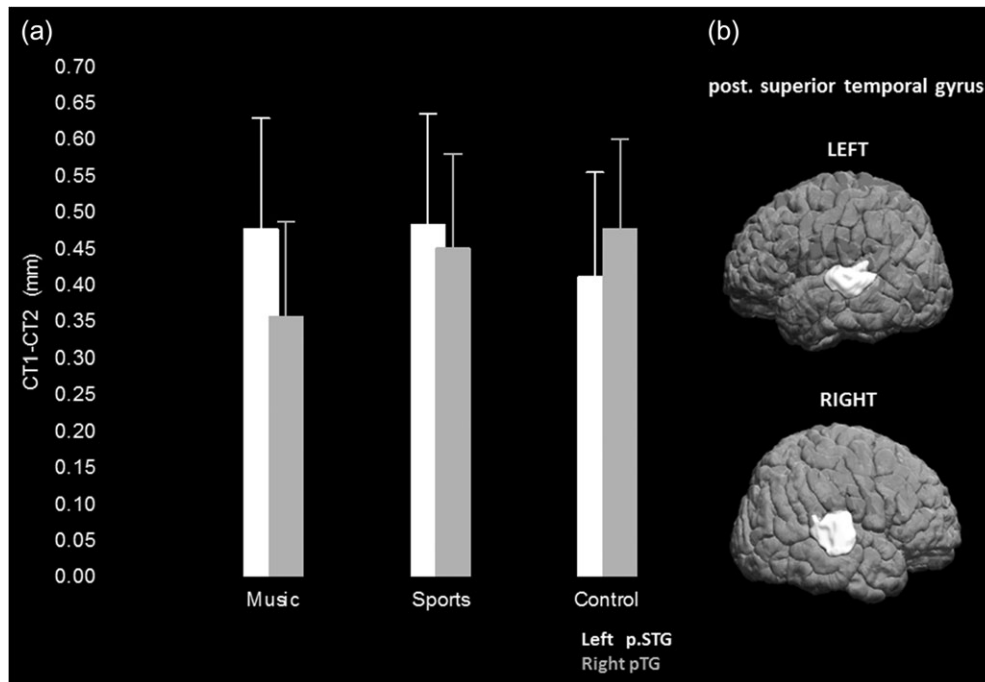


Figure 1. (a) Rate of cortical thinning on the left and right posterior superior temporal gyrus in music, sports and control groups. (b) Segmentation of the posterior superior temporal gyrus on the left and right hemispheres.

8. Posterior superior temporal gyrus: there was no significant group difference $F(2, 53) = 0.70, P = 0.49, \eta^2 = 0.02$. The decrease of CV was significantly larger on the left versus right posterior superior temporal gyrus as evidenced by significant main effect of laterality $F(1, 53) = 8.08, P = 0.006, \eta^2 = 0.13$. The group by laterality interaction reached marginal significance, $F(2, 53) = 2.44, P = 0.09, \eta^2 = 0.08$, with the music group showing greater (albeit not significant) CV loss on the left versus the right posterior superior temporal gyrus followed by the sports and control group (Music: Left vs. Right $P = 0.12$; Sports: Left vs. Right, $P = 0.17$ and Control, Left vs. Right $P = 0.99$).

Diffusion

Results from 9 participants (Music = 3, Sports = 3 and Control = 3) had to be excluded from the diffusion analysis due to motion related artifacts in the diffusion data or misalignment to the MPRAGE scan.

Fractional anisotropy

Scan 1. There was no significant difference between groups in the whole corpus callosum $F(2, 44) = 2.1, P = 0.12, \eta^2 = 0.09$; nor was there a significant difference between groups in any of the 7 identified segments: frontal.polaе.orbito $F(2, 44) = 1.6, P = 0.21, \eta^2 = 0.06$; SFG-SMA $F(2, 44) = 1.64, P = 0.2, \eta^2 = 0.06$; motor $F(2, 44) = 1.2, P = 0.28, \eta^2 = 0.05$; sensory $F(2, 44) = 1.84, P = 0.16, \eta^2 = 0.07$; temporal $F(2, 44) = 0.2, P = 0.81, \eta^2 = 0.009$; SPL $F(2, 44) = 2.03, P = 0.14, \eta^2 = 0.08$, and OC-Pre Cun $F(2, 44) = 0.5, P = 0.6, \eta^2 = 0.02$.

Scan 2. There was no significant difference between groups in the whole corpus callosum $F(2, 40) = 0.33, P = 0.71, \eta^2 = 0.01$ at scan 2; however, the FA was significantly different between groups in SFG-SMA segment $F(2, 40) = 4.19, P = 0.02, \eta^2 = 0.17$

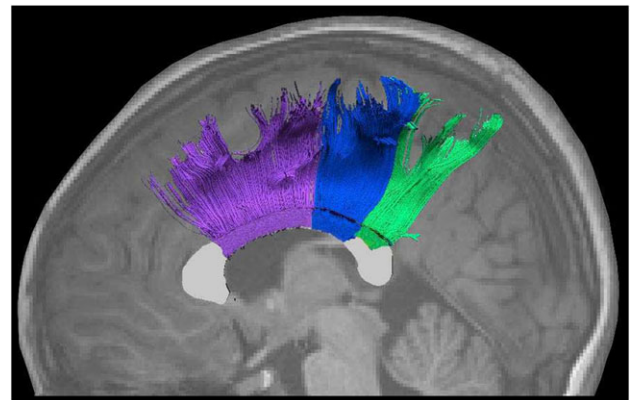


Figure 2. FA was increased at time of scan 2 in the music group, compared to two control groups, in the connections between superior frontal gyri (magenta), precentral gyri (blue) and the post-central gyrus (light green) of the corpus callosum.

($M > S: P = 0.01, M \text{ vs. } C: P = 0.32, S \text{ vs. } C: P = 0.32$); motor segment $F(2, 41) = 3.32, P = 0.04, \eta^2 = 0.14$ ($M > S: P = 0.03, M \text{ vs. } C: P = 0.23, S \text{ vs. } C: P = 0.63$); and marginally in the sensory segment $F(2, 40) = 2.81, P = 0.07, \eta^2 = 0.12$, of the corpus callosum (see Fig. 2); in all 3 cases the FA for the music group was higher compared to the values for the sports and control groups. No significant difference of FA was observed in the frontal.polaе.orbito $F(2, 40) = 1.79, P = 0.17, \eta^2 = 0.08$; temporal $F(2, 40) = 0.66, P = 0.51, \eta^2 = 0.03$ SPL $F(2, 40) = 1.36, P = 0.26, \eta^2 = 0.06$ or OC-Pre Cun $F(2, 40) = 1.45, P = 0.24, \eta^2 = 0.06$.

Mean Diffusivity

Scan 1. There was no significant difference of MD among groups in the whole corpus callosum $F(2, 44) = 0.04, P = 0.95, \eta^2 = 0.01$; neither in any of the segments of the callosum: frontal.polaе.

orbito $F(2, 44) = 2.31, P = 0.11, \eta^2 = 0.09$; SFG-SMA $F(2, 44) = 0.43, P = 0.65, \eta^2 = 0.01$; motor $F(2, 44) = 0.28, P = 0.75, \eta^2 = 0.01$; sensory $F(2, 44) = 0.28, P = 0.75, \eta^2 = 0.01$, temporal $F(2, 44) = 0.13, P = 0.87, \eta^2 = 0.006$, SPL $F(2, 44) = 0.46, P = 0.63, \eta^2 = 0.02$ and OC-Pre Cun $F(2, 44) = 0.4, P = 0.66, \eta^2 = 0.01$.

Scan 2. There was no significant difference among groups in the whole corpus callosum $F(2, 40) = 1.50, P = 0.23, \eta^2 = 0.07$; nor in any of the segments: frontal.polae.orbito-cc $F(2, 40) = 0.81, P = 0.44, \eta^2 = 0.03$; SFG-SMA $F(2, 40) = 0.99, P = 0.37, \eta^2 = 0.04$; motor $F(2, 40) = 0.03, P = 0.96, \eta^2 = 0.001$; sensory $F(2, 40) = 0.34, P = 0.71, \eta^2 = 0.01$; temporal $F(2, 40) = 0.187, P = 0.16, \eta^2 = 0.08$; SPL $F(2, 40) = 1.04, P = 0.36, \eta^2 = 0.04$, and OC-Pre Cun $F(2, 40) = 1.98, P = 0.15, \eta^2 = 0.09$.

Discussion

We investigated the impact of music training on the development of the brain in children ages 6–7, prior to their participation in music training and two years after the start of training. The analyses were performed at both macro- and micro-structural levels. We compared children involved in regular music training with 2 age-matched control groups (one of which involved systematic, regular athletic training), but not any music practice. All the three groups had the same low same socio-economic background and came from equally low-income areas of Los Angeles.

We report two main findings: (1) an asymmetric reduction of CT and volume of the posterior segment of the superior temporal gyrus (larger on the left than on the right) in the music group. This finding came from the comparison of scan 1 (prior to music training) to scan 2 (after 2 years of music training). No such asymmetric reduction was seen in either control group; although, all children showed some degree of CT reduction over the whole brain, as expected. (2) An increase in FA in 3 segments of the corpus callosum (CC) for the music group compared to the control groups after 2 years of music training; the segments showing differences among the groups correspond to the crossing of sensory, motor, and superior frontal gyri/supplementary motor area connections). These microstructural differences were not present at the time of the first scan. We will discuss each of these results separately and in relation to findings reported previously in the relevant literature.

Music Training and Development of Brain Auditory Regions

Our results indicate an overall decline in CT across the brain, in all three groups. This is consistent with reports showing that the decline in CT is greatest in childhood and follows a linear and gradual decrease, during adolescence and early adulthood. This maturation-related decline is not homogenous across varied brain regions (Mills and Tamnes 2014). For example, language areas have been reported to show thickening of the cortex during the period associated with fine tuning of language skills (Sowell et al. 2004). We believe it is reasonable to interpret the findings in our music group (between-hemispheric differences in the reduction of CT and volume in the posterior superior temporal gyri—auditory association regions) as an indication that the structural changes were related to musical training.

Our finding is in line with previous reports of music training-induced structural and functional brain differences between adult musicians and nonmusicians, specifically within

auditory regions (see Pantev et al. 2001; Schlaug 2001). Functional leftward asymmetry of the auditory cortices including the planum temporale, had been demonstrated as early as 1995 by Schlaug and colleagues as a function of musical training (Schlaug et al. 1995a). Schneider et al. (2002), reported that the gray matter volume of the anterior segment of the Heschl's gyrus was correlated with musical aptitude and was significantly greater in musicians compared to nonmusicians, (Schneider et al. 2002). Using a voxel-based morphometric (VBM) technique, Gaser and Schlaug reported a positive correlation between musical proficiency and gray matter density of the left Heschl's gyrus, when comparing professional musicians, amateur musicians and nonmusicians (Gaser and Schlaug 2003). Bermudez and colleagues, in the first study using multimodal CT and VBM measures to compare musicians and nonmusicians, reported greater CT and higher gray matter concentration in musicians, bilaterally, but especially noticeable in the right superior temporal surfaces corresponding to the planum temporale (Bermudez et al. 2009). Using surface-based morphometry, Elmer and colleagues showed that differences in cortical surface area of the planum temporale in musicians compared to nonmusicians, is associated with better processing of fast changing phonetic-cues in a phonetic categorization task (Elmer et al. 2013). And in 2012, local structural connectivity in peri-sylvian areas was found to be significantly increased, in the planum temporale, Heschl's and superior temporal gyri in musicians, particularly in those with absolute pitch abilities (Jäncke et al. 2012).

Given the cross-sectional nature of the above studies, which were carried out in adults, pre-existing genetic dispositions and the environment may have contributed to the findings. Hyde and colleagues, in the first longitudinal study of structural changes related to the learning of a musical instrument demonstrated that 6-year-old children receiving instrumental musical training for 15 months showed increased gray matter density in the right primary auditory cortex (Heschl's/transverse temporal gyrus) while age-matched children receiving no musical training did not (Hyde et al. 2009a).

Our results revealed that in children in the music group relative to the two control groups, a slower rate of cortical thinning took place in the right posterior segment of the superior temporal gyrus than in the left. With respect to CVs, changes in the same region followed a similar pattern (although the change remained at the level of a strong trend that did not quite meet significance). In contrast to the results of Hyde et al. (2009a), the music training-related structural difference we observed is outside of the primary auditory cortex, in the posterior segment of the superior temporal gyrus, an area that has been shown to have an important role in auditory feature extraction and the processing of complex sounds including music (Koelsch et al. 2005; Koelsch 2011). Discrepancies in findings may reflect differences in methods of analysis (surface-based cortical reconstruction versus VBM approaches), differences in the participants, and even differences in the type of music training itself. For example, the VBM techniques used by Hyde and colleagues, provides a comparison of gray matter density on a voxel-wise basis, after fitting of individual brain volumes to a common template. This registration process can jeopardize the accuracy to localize region-specific changes in the brain structure (Bookstein 2001). Furthermore, results of VBM analysis are typically referred to as gray matter concentration or gray matter density, which does not have a biological correspondence and therefore is not easy to evaluate in the context of brain maturation in a longitudinal setting. We used a surface-based cortical reconstruction method

that identifies the border between tissue types (gray matter, white matter, and CSF) and permits assessment of not only CV but also CT, a measure that may more closely reflect cytoarchitectural properties than gray matter density. Lastly, unlike VBM, each ROI measure was obtained in the actual brain-space of each individual. Of the 16 ROIs studied for gray matter thickness change (8 on each side) only the posterior segment of the superior temporal gyrus (pSTG) showed a different rate of cortical thinning. The music group specifically showed a difference between the CT change of the pSTG on the right versus left hemisphere consistent with a lower right-sided cortical thinning relative the left-sided thinning. This rightward asymmetry in our findings of a lesser reduction of CV and thickness of this region is well-supported by previous reports of functional lateralization, namely a rightward asymmetry, of auditory association areas, engaged in processing of complex auditory stimuli including music (Hyde et al. 2008; Bermudez et al. 2009; Burke 2010).

Hudziak and colleagues, in a recent cross-sectional study, have showed that music training was associated with differences in CT (which they attributed to an increased developmental thinning of cortical areas) in children ages 10 and older, within areas related to motor planning, coordination and visuospatial abilities (Hudziak et al. 2014). No association was reported in this study between music training and changes in primary or secondary auditory areas. The normal course of cortical maturation has been shown to be associated with an overall decline in CT (Sowell et al. 2004). However, the maturation rate is not homogenous across different brain regions; first to mature are sensory and motor regions that are involved with more basic functions and last are higher order association areas that are involved with integration of attention, memory and executive function (Gogtay et al. 2004). Within the temporal lobe, specifically, the posterior superior temporal gyrus has been shown to mature late in development and to continue to develop into early adulthood (Gogtay et al. 2004). Therefore at different stages of development, the pattern of relationship between a behavioral outcome and cortical growth may be different. For example, using a longitudinal design, Shaw and colleagues (2006), identified a marked developmental shift from a predominately negative correlation between intelligence and CT in early childhood (ages 3–8) to a positive correlation in late childhood and beyond (ages 9–12).

Children in our study were first scanned between ages 6–7 and again between ages 8–9. An overall decline in CT was noted in all groups and in all ROIs; however it appears that changes related to music training, specifically within the right posterior segment of superior temporal gyrus, is marked by a possible experience-dependent increase in CT—rather than a decrease—related to the frequent and systematic engagement of the right pSTG in musical training processes. Consequently, we interpret the reduction in the rate of cortical thinning on the right versus left pSTG, as related to the interaction of the normal course of cortical thinning of auditory association areas; with the auditory stimulation induced by early music training, which would influence the change in CT in the opposite direction. This interpretation would be concordant with the experience-based thickening of cortex previously reported in language related areas in association with fine tuning and mastery of linguistic skills during late childhood (5–11) (Sowell et al. 2004). Positive correlations have been shown between the trajectory of CT maturation both with IQ (Shaw et al. 2006) and, separately, with time playing a musical instrument (Hudziak et al. 2014) during the developmental stage of late

childhood (ages 8–10). Our results, therefore, suggest that early exposure to music training may be related to the long-term effects of increased CT of auditory areas previously reported in adult musicians (Bermudez et al. 2009).

The biological mechanisms accounting for the differences we report here cannot be determined from our data. It is possible, however, that while early brain development is characterized by overproduction of synapses and a parallel pruning process, experience-based neural activity—corresponding in our case, to recurring neural activation in the auditory regions related to playing music—can guide the pruning process and determine which synaptic connections are preserved (Stoneham et al. 2010). This would result in changes in the morphology of auditory regions. Other biological mechanisms can possibly account for the changes observed here. This would include experience-dependent synaptogenesis or increased number of glial cells associated with learning (Zatorre et al. 2012; Bailey et al. 2014). Independently of the mechanism underlying the structural changes we report here, they happened in parallel with functional improvements such as the enhanced ability to detect changes in tonal environment (see Ilari et al. 2016) and the accelerated maturity of auditory processing, as evidenced by an accelerated development of adult-like cortical auditory evoked potentials (see Habibi et al. 2016). Such behavioral and electrophysiological changes have also been reported by Tierney et al. (2015). Enhanced maturation of these auditory abilities may also lead, perhaps later, to faster and more efficient development of language, speech and auditory memory skills, given that some of the neural substrates of these different processes are in fact shared.

Music Training and Microstructural Development of the Corpus Callosum

We showed that children with 2 years of music training have larger FA in the corpus callosum (CC), specifically at the level of crossing fibers connecting superior frontal, sensory and motor segments across the callosum, compared to the children in the two control groups. The differences between music and sports groups were significant; however, the differences between music and control did not reach significance. Given that none of the differences were present at the first assessment (all $P > 0.1$), prior to any training, we suggest that the musical experience may have played a role in the microstructural changes detected in the white matter of the corpus callosum.

Maturation of the white matter has been related to measures of diffusion imaging variables, including MD, which reflects overall magnitude of water diffusion and FA, which indexes degree of net directionality in water diffusion in the tissue. Widespread white matter development across the brain, during childhood and adolescence, has been associated with FA increases and MD decreases, although regional differences exist (Mills and Tamnes 2014). Maturation of corpus callosum as the main interhemispheric region of crossing of fiber tracts that integrate communication between the two hemispheres is indexed by increase in overall size and increase in FA value. This process begins in early childhood and has been shown to continue to the second decade of adulthood (Pujol et al. 1993; Westerhausen et al. 2011).

In an early study, measuring the size of CC and its subdivisions in adult musicians and matched control participants, Schlaug and colleagues reported a larger anterior CC in musicians compared to nonmusicians, and that the difference was greater for musicians with an early onset of training, before age 7. These findings were interpreted as indicating increased

interhemispheric communication in the premotor and supplementary motor areas of the frontal cortices in musicians, possibly related to the complex bimanual motor coordination demanded in playing a music instrument (Schlaug et al. 1995b). Using diffusion imaging, Schmithorst and Wilke, compared adult musicians with nonmusicians, and reported that musicians have greater FA in the genu of the corpus callosum (Schmithorst and Wilke 2002). Furthermore, comparing white matter parameters in musicians with early and late age onset of music training to nonmusicians, Steele and colleagues found that musicians with early onset of training had greater connectivity and increased FA in the posterior midbody/isthmus segment of the corpus callosum containing fibers that connect sensorimotor cortices of the two hemispheres (Steele et al. 2013). Finally, in the report by Hyde and colleagues mentioned earlier, and consistent with our findings, the 6–7-year-old children showed increased volume in a similar region of the CC connecting sensory and motor regions after only 15 months of musical training (Hyde et al. 2009a).

Playing a musical instrument requires processing of sound, coordination of both hands and integration of auditory and motor demands. This may place greater demands on interhemispheric interaction between sensorimotor regions and in turn promote maturation of the pathways that connect these regions. Such maturation may be reflected in the increase of FA value in these segments of the CC. These microstructural differences may reflect variation in white matter features, such as degree of axonal myelination (Alexander et al. 2007) which may be accelerated as a result of enhanced interaction between the auditory and motor regions across the 2 hemispheres; however, the exact underlying mechanism cannot be established on the basis of the current data.

Divergence from Other Studies

Contrary to previous findings (Hyde et al. 2009b; Bailey et al. 2014), we did not observe any CT or volume differences between the music and control groups outside of the auditory areas (e.g., in the inferior frontal gyrus). Because we are investigating brains of young children during a period of intense change related to typical development, the short time of intervention about 2 years (23–25 months), and the limited intensity of the intervention possibly explain the lack of more widespread differences. The fact that we detected discriminating changes in the auditory regions and in the corpus callosum seems to indicate that there was an effect on regions directly stimulated by the intervention and suggests that, differences in functionally related regions may well appear at a later stage. Structural-dependent changes have been shown to correlate with behavioral changes; as, for example, performance on an auditory-motor synchronization task shown to correlate with increased gray matter concentration of the ventral premotor cortex (Bailey et al. 2014). We observed improvements in musical skills such as tonal perception after only 2 years, and these were limited to the music group (Habibi et al. 2016); however, significant group differences in rhythm perception or auditory-motor synchronization were not detected at the time of second scan (Ilari et al. 2016).

Limitations of the Current Study

An inevitable limitation of our study is the fact that it was not based on a randomized controlled trial design. Albeit unlikely, it is not possible to rule out that musicians may be born with pre-existing biological tendencies for musicality, or that some of the children may have had a certain biological predispositions to

brain changes related to musical training, or have had parents who were motivated to enroll them in music training in the first place and therefore the possibility of a different home environment with respect to music. Our groups were formed of typically developing children who enrolled in their respective extracurricular programs (music or sports) by their (and their family's) motivation. On the other hand, if children were not motivated and not emotionally engaged in the chosen activity, it is unlikely that they would continue participation over the long period necessary for a longitudinal investigation. In addition, assigning children to specifically not engage in an activity thought to be beneficial during critical times of development, and for long periods, would simply be unethical. Randomized trials are ideal and feasible for short-term clinical interventions. It would be beneficial if such a study design could be ethically and practically developed for the study of systematic musical training but we cannot see it happening in the near future. In the meantime, "real world" studies such as ours, can help develop a deeper understanding of how music training may shape brain development in children. In addition, it can show the results of such training in an underprivileged socio-economic population without access to music education and therefore highlight the importance of inclusion of such programs within childhood education curricula.

Another limitation can be seen in the relative small number of participants. This was the result of significant logistical challenges in recruiting and retaining participants; especially in a study involving children from low socio-economic backgrounds. Under the circumstances, we were limited to 25 children in the music group recruited over a 2-year period and we attempted to match the numbers in the 2 control groups. Given that the 3 groups showed no significant differences on any assessed measure of cognitive, social, emotional, or brain structure and function at the time of induction (all $P > 0.1$; see Habibi et al. 2014), and the effect sizes for the observed differences after two years of training were of moderate to large size (range of $\eta^2 = 0.12$ – 0.17), we believe the results reported here provide a significant contribution to the understanding of the important role of music training in the brain development of children. We also note that individually these results do not survive a multiple comparison correction (i.e., Bonferroni), possibly because of the limited sample size. However, this is mitigated, to some degree, by the consistency in findings between CT and ROI volume in the posterior segment of the superior temporal gyrus, and by the fact that we found significant (uncorrected) effects in 3 of 7 subdivisions of the corpus callosum. Finally, the results reported here are part of an ongoing longitudinal study with one further neuroimaging data collection session scheduled to take place 2 years after the collection of the findings reported here, bringing the total music training period to 4 years. Given that length of music training has been previously shown to correlate with changes in brain morphology in adult musicians (Bengtsson et al. 2005; Hudziak et al. 2014), we may expect, at that point, a larger difference between the music and the two control groups, as well as involvement of further brain regions.

In conclusion, we have documented longitudinal changes in brain development at macro-structural and microstructural levels, distinct from the typical brain changes seen during development. These changes were shown in young children from underprivileged communities who underwent 2 years of musical training; we have also showed that these macro-structural and microstructural changes were distinct from those seen in 2 age-matched control groups from similar backgrounds (one of which underwent an equally motivating intervention, which was comparable in sensory-motor demands but did not

have any musical involvement). To the best of our knowledge this is the first study with these characteristics. The regional, training-related structural and microstructural changes we found were present in musically-relevant brain regions, including association auditory regions and the corpus callosum. These changes, given that there were no brain differences among the groups prior to the onset of the study provide strong evidence that the differential rate of maturation of these brain regions is probably related to music training rather than to pre-existing biological dispositions for musicality. Our findings also suggest that music training is a powerful intervention that may facilitate regional brain maturation during childhood.

Supplementary Material

Supplementary data are available at *Cerebral Cortex* online.

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Notes

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