Classification: Biological Sciences; Neuroscience

Title: Adolescence is associated with genomically patterned consolidation of the hubs of the human brain connectome

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Abstract

How does human brain structure mature during adolescence? We used MRI to measure cortical thickness and intra-cortical myelination in 297 population volunteers aged 14-24 years. We found, and replicated, that association cortical areas were thicker and less myelinated than primary cortical areas at 14 years. However, association cortex had faster rates of shrinkage and myelination over the course of adolescence. Age-related increases in cortical myelination were maximised approximately at the internal layer of projection neurons. Adolescent cortical myelination and shrinkage were coupled and specifically associated with a dorso-ventrally patterned gene expression profile enriched for synaptic, oligodendroglial and schizophrenia-related genes. Topologically efficient and biologically expensive hubs of the brain anatomical network had greater rates of shrinkage/myelination, and were associated with over-expression of the same transcriptional profile as cortical consolidation. We conclude that normative human brain maturation involves a genetically patterned process of consolidating anatomical network hubs. We argue that developmental variation of this consolidation process may be relevant both to normal cognitive and behavioural changes, and to the high incidence of schizophrenia, during human brain adolescence.

Significance Statement

Adolescence is a period of human brain growth and of high incidence of mental health disorders. Here we show, consistently in two MRI cohorts, that human brain changes in adolescence were concentrated on the more densely connected hubs of the connectome – i.e., association cortical regions that mediated efficient connectivity throughout the human brain structural network. Hubs were less myelinated at 14 years, but had faster rates of myelination and cortical shrinkage in the 14–24 year period. This topologically focused process of cortical consolidation was associated with expression of genes enriched for normal synaptic and myelin-related processes, and for risk of schizophrenia. Consolidation of anatomical network hubs could be important for normal and clinically disordered adolescent brain development.

Main Text

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Adolescence is associated with major behavioral, social and sexual changes, as well as increased risk for many psychiatric disorders (1). However, human brain maturation during adolescence is not yet so well understood. Historically pioneering studies used histological techniques to demonstrate that distinct areas of cortex were differentially myelinated in post mortem examination of perinatal tissue, suggesting "myelinogenesis" as an important process in human brain development (2, 3). Magnetic resonance imaging (MRI) can measure human brain development more comprehensively, and over a wider age range, than is possible for post mortem anatomists. The thickness of human cortex can be reliably and replicably measured by MRI (4) and longitudinal studies have shown that cortical thickness (CT, mm) monotonically shrinks over the course of post-natal development with variable shrinkage rates estimated for different age ranges (5–11; for review see 12). CT typically shrinks from about 3.5 mm at age 13 years (9) to about 2.2 mm at age 75 (10, 11). Rates of cortical shrinkage are faster during adolescence (approximately -0.05 mm/year) than in later adulthood or earlier childhood (9).

What does this MRI phenomenon of cortical shrinkage represent at a cellular level? There are broadly two tenable models: pruning and myelination. Basic physical principles of MRI predict that shorter longitudinal (T1) relaxation times reflect either a reduction in the fraction of "watery" cytoplasmic material, like cell bodies, synapses or extra-cellular fluid; or an increase in the fraction of "fatty" myelinated material, like axons. Pruning models propose that cortical shrinkage in adolescence represents loss or remodeling of synapses, dendrites, or cell bodies (13). Myelination models propose that the cortex appears to shrink due to an increasing proportion of myelinated axons, without necessarily implying any loss or change of neuronal material (5).

In the macaque monkey, although the main phase of synaptic pruning and neuronal loss occurs earlier in development (14, 15), there is evidence for further synaptic remodeling during adolescence (16, 17). In rodents, there is histological evidence for increasing intra-cortical myelination during adolescence, especially at the deeper cytoarchitectonic layers of cortex (V and VI; 18, 19). At a cellular level, this likely reflects a developmentally late myelination of

efferent axonal segments immediately distal to the axonal hillock of pyramidal cells (20). MRI sequences have been recently developed for myelin mapping in humans (21). These techniques include methods based on magnetisation transfer (MT) which have been validated as an MRI marker of myelination by post mortem imaging and histological studies of multiple sclerosis, a demyelinating disorder (22); see **Methods** for details. It has been shown that primary sensory and motor areas of cortex are more heavily myelinated than association cortical areas; that most cortical areas show progressive increases in myelination over the course of normative development into middle age; and that myelination is concentrated in deeper layers of cortex (21, 23, 24). Association cortical areas have been identified among the highly connected "hubs" of structural brain networks (25), suggesting that differential rates of intracortical myelination might be related to differences between regions in their topological roles as part of the connectome. Developmental changes in structural and functional MRI network topology have been reported (for review see 26) but not previously related to measures of cortical shrinkage or myelination. Recently, several studies have linked brain regional gene expression to axonal connectivity in the mouse (27, 28) and to functional MRI networks in humans (29, 30), but there have been no previous efforts to investigate genetic mechanisms of adolescent myelination of human cortex.

In this context, we aimed to test three hypotheses: i) that adolescent cortical shrinkage was coupled to intra-cortical myelination; ii) that adolescent cortical shrinkage/myelination, a.k.a. consolidation, was concentrated anatomically on association cortex and topologically on the most strongly connected regions (hubs) of the human brain anatomical network; and iii) that adolescent consolidation of these connectome hubs was associated with a specific gene expression profile, enriched for neuronal and oligodendroglial function; and enriched for risk of schizophrenia, a neurodevelopmental disorder with its highest incidence in young adults.

We used quantitative multi-parameter mapping (MPM; 31) to test these hypotheses on MRI data acquired from a sample of 297 healthy young people sampled from primary health care registers, stratified by age, and balanced for sex in the adolescent age range 14-24 years, with approximately 60 participants in each of 5 age-defined strata: 14-15 years inclusive, 16-17 years, 18-19 years, 20-21 years, and 22-24 years (see **Methods** and **Table S1**). From the MRI data, we measured cortical thickness (CT, mm) and magnetisation transfer (MT, percentage units [PU]) at

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each of 308 cortical regions for all participants. We used linear models to estimate baseline CT and MT at 14 years, and age-related rates of change in the period 14-24 years (ΔCT, ΔMT), from data on participants of all ages at each regional node. We explored the relationships between these local cortical MRI markers and a few, key metrics of complex network topology that have been widely used in prior neuroimaging and other neuroscience studies (for review see 32). We focused on the degree and closeness of each node - as measures of nodal "hubness" - and the community structure of the network - defined as a set of sparsely inter-connected modules; see **Methods** and **Supplementary Information (SI)** for details of topological connectome analysis. We investigated the relationships between gene transcriptional profiles and co-localised MRI (CT, MT) and network topological phenotypes by multivariate analysis of MRI data on 297 adolescents and whole-genome gene expression maps of 6 adult human brains (post mortem) provided by the Allen Institute for Brain Science (AIBS; 33); see **Methods** and **SI** for details.

For robustness and generalizability, we first analyzed data from a discovery cohort (N=100; balanced for age and sex as per the sample design) and then replicated all the key findings in a validation cohort (N=197). Non-identifiable data and all analysis code are available at the NSPN Cortical Myelination figshare repository (see **SI** for details).

<u>Results</u>

Cortical thickness and shrinkage

Cortical thickness at 14 years of age ranged between 1.93 and 3.8 mm across different cortical areas. Baseline CT was thinnest in primary somatosensory and visual cortices (**Fig. 1a**; 34). In the adolescent period from 14-24 years, there was evidence for significant cortical shrinkage (r^2 = 0.10, P = .006, estimated global rate of shrinkage, ΔCT = -0.011 mm/year; **Fig. 1b**). Although 289 of the 308 nodes exhibited cortical shrinkage (ΔCT < 0), only 79 nodes showed shrinkage that was statistically significant at P < .05 after FDR correction for multiple comparisons (**Fig. 1c**). The regions with the greatest rates of shrinkage were located in association cortex.

Intra-cortical magnetisation transfer and myelination

By combining data from neighbouring voxels in each regional node, we could estimate regional MT for each depth at sub-millimetre resolution (4), corresponding approximately to different

layers of cortex (**Fig. 2a**). MT was estimated at ten fractional depths of the cortical ribbon from the pial surface (0% depth) to the grey/white matter boundary (100% depth), and at two infracortical locations 0.4mm and 0.8mm below the grey/white matter boundary. At each cortical depth, we estimated baseline MT (at age 14) and adolescent change in MT (over the age range 14-24 years). In all regions, as expected, baseline MT increased monotonically as a function of increasing depth from the pial surface, reflecting greater density of myelinated fibres in deeper cortical layers (**Fig. 2b**; **Movie S1**). However, there was marked regional variability (**Fig. 1c**): areas of primary sensory cortex had greater MT at age 14 than association cortical areas.

Rates of adolescent change in MT, ΔMT, were greatest when measured at 70% of the depth of cortex (**Fig. 2d**; **Movie S2**), i.e., ~1.9 mm below the pial surface and ~0.8 mm above the boundary with white matter (across all cortical regions: $r^2 = 0.17$, $P < .001$, ΔMT $= 4.98 \times 10^{-3}$ year^{-1}; **Fig. 1d**). 70% cortical depth corresponds approximately to the level of lamina V and VI, comprising the internal layer of pyramidal or projection neurons (**Fig. 2c**). There was marked regional variability in the rates of myelination: regions with the greatest rates of myelination were located in association cortices (**Fig. 1g**).

In light of these complementary anatomical and developmental profiles of CT and MT, it is not surprising that there was a strong negative correlation between baseline cortical thickness and magnetisation transfer ($r^2 = 0.43$, $P < .001$, $\beta = -0.120$ PU/mm; **Fig. 1f**) and a strong negative correlation between rates of cortical shrinkage and intra-cortical myelination ($r^2 = 0.22$, $P < .001$, $\beta = -0.126$ PU/mm; **Fig. 1h**). Adolescent cortical shrinkage was significantly but not entirely mediated by age-dependent changes in MT (**Fig. S1E**), indicating that myelination is necessary but not sufficient to account for cortical shrinkage.

Internal replication

All of the results reported so far were based on analysis of the discovery cohort (N=100; 20 in each of 5 age bins; 50 female); and all were closely replicated in the validation cohort (N=197; ~40 in each of 5 age bins; 98 female; **Figs. S1** and **S2**). As the validation cohort had twice as many participants, it conferred greater statistical power to test hypotheses. We henceforth focus on the most precise estimates of CT and MT (at 70% cortical depth) estimated from the total

sample (N=297; 60 per age bin, 30 female), although all further analyses were also reproduced separately for the discovery and validation cohorts (**Figs. S3** and **S4**).

Genomic patterning of adolescent cortical consolidation

To elucidate the molecular mechanisms of local change in CT and MT, we explored the association between these MRI markers and regional gene expression profiles from human adult brain microarray datasets provided by AIBS (33). For example, baseline MT was correlated with regional expression of the gene for myelin basic protein (*MBP*) across cortex (**Fig. 2e**) confirming that the MT signal is indicative of myelin content (22). The association was strongest at 70% cortical depth ($r^2 = 0.21$, $P < .001$; $\beta = 0.0723$; **Fig. S3A**).

To explore analogous associations between all 4 cortical MRI metrics, and all 20,737 genes measured in the AIBS microarrays, at each of 308 regions, we used the multivariate, dimension-reducing technique of partial least squares (PLS; 35). This analysis defined a few PLS components which were the linear combinations of the weighted gene expression scores (predictor variables) that were most strongly correlated with one or more of the MRI markers (response variables; CT14, MT14, ΔCT and ΔMT).

The top two PLS components explained 28% of the variance in the MRI response variables (permutation test, $P < .001$). The first PLS component (PLS1; **Fig. 3a**) represented a significant association between a rostro-caudally patterned gene expression profile and baseline measures of CT and MT at 14 years (**Fig. 3b-c**; **Fig. S5**). The second, independent, PLS component (PLS2; **Fig. 3d**) represented a significant association between a dorso-ventrally patterned gene expression profile and measures of adolescent cortical shrinkage and myelination (**Fig. 3e-f**). See **Supplementary File 1** for a full list of significantly over- or under-expressed genes represented by the first two PLS components.

Focusing on the gene expression profile defined by the second PLS component, because it was specifically associated with adolescent cortical shrinkage and intra-cortical myelination, we found that this transcriptional signature was significantly enriched in genes relating to synaptic transmission ($P < .001$), regulation of glutamatergic signaling ($P < .001$), and potassium ion channels ($P < .001$; **Fig. 3g**; 36). We also found that the transcriptional profile associated with

association cortical consolidation was significantly enriched for an oligodendroglial gene set (P < .001; **Fig. S3F**; 37), as well as for a set of genes robustly associated with risk for schizophrenia, a neurodevelopmental disorder (P < .001; **Fig. S3G**; 38).

Adolescent cortical consolidation and the connectome

We used the measurements of cortical thickness on the total sample to estimate the mean structural covariance matrix, representing the pair-wise correlation of cortical thickness between each possible pair of 308 regions. This matrix was thresholded to construct a binary graph or structural covariance network which had a complex topology, consistent with many prior reports of human anatomical connectomes (**Fig. S6**; 39).

Globally, the network was small-world with hubs (indicated by a fat-tailed degree distribution), a modular community structure, and a rich club or core of densely inter-connected high degree nodes or hubs, which were located primarily in frontal and parietal association cortices (**Fig. 4a**, **Fig. S6**). At a nodal level of analysis, high degree hub nodes also had high closeness centrality, indicating short path length of connections to other nodes in the network (**Fig. 4b**), as well as long physical distance of connections. Cortical shrinkage was faster for topologically central hubs with long distance connections (**Fig. 4c**; degree: $r^2 = 0.14$, $P < .001$, $\beta = -1.48 \times 10^{-3} \text{mm}^{-1}$; closeness: $r^2 = 0.18$, $P < .001$, $\beta = -4.91 \text{ mm}^{-1}$; distance: $r^2 = 0.09$, $P < .001$, $\beta = -0.81 \times 10^{-3}$),. Intra-cortical myelination rates were also faster for long-distance hubs (**Fig. 4d**; degree: $r^2 = 0.07$, $P < .001$, $\beta = 3.81 \times 10^3 \text{ PU}^{-1}$; closeness: $r^2 = 0.13$, $P < .001$, $\beta = 14.8 \text{ PU}^{-1}$; distance: $r^2 = 0.21$, $P < .001$, $\beta = 4.42 \times 10^3 \text{ mm/PU}$).

We found that degree, closeness and connection distance were also positively correlated with the pattern of gene expression associated with adolescent change in cortical structure (PLS2; **Fig. 4e**; degree: $r^2 = 0.21$, $P < .001$, $\beta = 178$; closeness: $r^2 = 0.26$, $P < .001$, $\beta = 572 \times 10^{-3}$; distance: $r^2 = 0.15$, $P < .001$, $\beta = 100 \text{mm}$). In contrast, these metrics were not so strongly correlated with the pattern of gene expression associated with cortical structure at 14 years (PLS1, degree: $r^2 < 0.01$, $P = .904$, $\beta = -2.36$; closeness: $r^2 < 0.01$, $P = .892$, $\beta = 9.39 \times 10^{-3}$; distance: $r^2 = 0.02$, $P = .02$, $\beta = -35.2 \text{mm}$).

<u>Discussion</u>

We have shown that adolescent cortical shrinkage is related to changes in intra-cortical magnetisation transfer in humans. This observation supports myelination models of shrinkage, and more generally confirms association cortical myelinogenesis as a key neurodevelopmental process in adolescence. Our baseline MT-based myelination maps (**Figs. 1** and **2**) show a high degree of correspondence with previous myelin maps based on alternative MRI parameters (23, 24, 40–42). Across modalities, the highest levels of myelin are located in areas of primary isocortex comprising many large pyramidal cells which are the principal targets and sources of axonal projections (43). However, arguably the strongest evidence that MT is representative of myelin content in these data is that MT was highly correlated with regional expression of the gene for myelin basic protein, and the set of genes most strongly correlated with adolescent change in MT was significantly enriched for oligodendroglia-related genes.

We interpret our findings as indicating that adolescent cortical myelination was greatest in association areas, which were least myelinated at age 14 years. It is plausible, though not directly demonstrated by these data, that specialised motor and sensory cortex may show faster rates of intracortical MT change compared to association cortex at earlier stages of development. This would be compatible with the idea, dating back to Flechsig (2), that association cortical areas are the focus of a relatively late wave of myelinogenesis. The intra-cortical location of strongest maturational changes in MT, corresponding approximately to the boundary between layer V (internal pyramid) and layer VI, further suggests that adolescent intracortical myelination may be concentrated on the proximal segments of efferent projections from pyramidal neurons (20).

Although intra-cortical myelination was greatest in areas that showed fastest rates of cortical shrinkage, it did not entirely explain age-related changes in cortical thickness. Likewise, although the gene expression profile associated with adolescent change was enriched for genes related to oligodendroglial function, it was also significantly enriched for neuronal genes, especially those implicated in remodeling of synapses or transport of glutamate-containing vesicles. Thus it seems plausible that genetically programmed processes of synaptic remodeling act together with locally coupled processes of intra-cortical myelination to consolidate synaptic and axonal connectivity of association cortical areas in adolescence. The set of gene transcription markers most strongly associated with this late maturational process (44) was enriched for genes known to confer risk for schizophrenia (38), generating the hypothesis that deviation from the

normative developmental trajectory of cortical hub consolidation could be an intermediate phenotype underlying the high incidence of the clinical phenotype of schizophrenia in young people.

However, we note some important caveats that mandate further critical testing of these results and their hypothetical implications. First, the gene expression profiles used in this analysis were measured in 6 post-mortem adult brains (mean age = 43 years). Brain regional gene expression is known to change somewhat over the two decades approximately intervening between the oldest subject in the MRI dataset and the average age of the subjects in the genomic dataset (45). Any such age-related changes in gene expression will have confounded our analysis of the association between imaging and genomic variables. We might expect this age disparity to reduce the statistical power to detect true MRI/mRNA associations, rather than to inflate the probability of false positive associations. Nonetheless, the genomic analysis of developmental MRI phenotypes would certainly be stronger in future if it was informed by age-matched regional gene expression profiles (such data are not currently available). Second, the MRI changes we have interpreted developmentally are estimated from cross-sectional data. A longitudinal design incorporating multiple repeated MRI scans over time would provide a more secure basis for estimation of age-related changes attributable to the developmental maturation of individual brains. Third, we have not presented any evidence that people at risk for schizophrenia do indeed demonstrate abnormal cortical hub consolidation, although we note there is prior evidence for abnormal cortical thickness, magnetisation transfer and cortical shrinkage in patients with schizophrenia (46–48). Future longitudinal studies of network development (and ideally also gene expression) in young people genetically or environmentally at risk for schizophrenia will be required to test the pathogenic role of hub consolidation more definitively.

It is well known that brain networks generally have complex topological properties, including hubs, modules and rich clubs (39, 49). We replicated these findings by graph theoretical analysis of structural covariance networks (derived from the CT measurements), showing that topologically central nodes had higher rates of adolescent shrinkage and myelination, and higher levels of gene expression associated with cortical consolidation. In a sense this is not surprising, given our earlier results showing that the coupled changes in CT and MT were greatest in association cortex, and prior studies showing that network hubs of the structural brain

connectome are disproportionately located in association cortex (39). It suggests that adolescent cortical consolidation is topologically targeted to optimise the performance of network hubs. For example, synaptic remodeling and intracortical myelination of cortical hubs might be expected to minimise the conduction time for axonal propagation of electrical signals or to enhance the synchronization of oscillations across anatomically distributed cortical areas (50). These physiological implications of connectome hub consolidation would hypothetically favour more integrative functional network topology and dynamics, which are known to be important for later-maturing, "higher order" cognitive functions (51).

We predict that age-related cortical consolidation of human cortical network hubs will prove to be relevant to the normal acquisition of cognitive and behavioural skills during the adolescent transition from childhood to adulthood, and may also prove to be implicated in the clinical emergence of neurodevelopmental and psychiatric disorders such as schizophrenia in young people (1, 3).

Methods

Design

2135 healthy young people in the age range 14-24 years were recruited from schools, colleges, NHS primary care services and direct advertisement in north London and Cambridgeshire. To populate the (secondary) MRI cohort, 300 participants were sub-sampled from the primary cohort (conserving sex and ethnicity balance) with N=60 in each of the 5 age strata. See **SI** for additional detail and exclusion criteria. Participants provided informed written consent for each aspect of the study and parental consent was obtained for those aged 14-15 years. The study was ethically approved by the National Research Ethics Service and was conducted in accordance with NHS research governance standards.

MRI data acquisition

All scans were acquired using the MPM sequence (31) implemented on three identical 3T whole body MRI systems (Magnetom TIM Trio, Siemens Healthcare, Erlangen, Germany; VB17 software version), two located in Cambridge and one in London. Between-site reliability and tolerability of all MRI procedures was satisfactorily assessed by a pilot study of 5 healthy volunteers each scanned (for approximately 25 mins) at each site (31). The between-site bias was less than 3%, and the between-site coefficient of variation was less than 8%, for both R1 and MT parameters (31). R1 and MT were quantified in Matlab (The MathWorks Inc., Natick, MA, USA) using SPM8 (www.fil.ion.ucl.ac.uk/spm) and custom tools; see **SI** for details.

Estimation of regional cortical thickness and MT

The cortical surface for each participant was reconstructed from their R1 image by the following steps: skull stripping (52), segmentation of cortical grey and white matter (53), separation of the two hemispheres and subcortical structures (53–55); and finally construction of smooth representation of the grey/white interface and the pial surface (53). We used FreeSurfer v5.3.0 software to implement these processes (http://surfer.nrm.mgh.harvard.edu). After quality control, three participants had to be excluded from the analyses due to movement artefacts which prevented accurate surface reconstructions. We used a backtracking algorithm (56), to parcellate the 66 regions defined by sulco-gyral criteria in the Desikan-Killiany atlas (57) into 308

contiguous parcels of approximately equal area ($500mm^2$) across both hemispheres in standard space (**Fig. S2A**). This parcellation template was then transformed to the native space of each participant's image to minimise geometric deformation of the MRI data and prevent age-related bias (58) prior to estimation of cortical thickness of each region. To localise cortical myelination at a spatial resolution corresponding approximately to the scale of the six lamina of neocortex, at each regional node, we estimated MT at each of 11 equidistant points on the normal line from the grey/white matter boundary (fractional cortical depth = 100%) to the pial surface (0%). By measuring MT at depths defined in proportion to cortical thickness, we aimed to adjust the measurements for variation in absolute cortical thickness between different regions and as a function of age. We additionally estimated MT at two locations, consistently defined as 0.4 mm and 0.8 mm below the GM/WM boundary, to sample the dense myelination of central white matter as a point of comparison for intracortical MT (**Fig. 2** and **SI** for detail).

Gene expression dataset

Microarray data for 5 male and 1 female donors with mean age 42.5 years were available from the Allen Institute for Brain Science (33; http://human.brain-map.org). We matched the centroids of the regions of the MRI parcellation to the closest regional gene expression profile. Microarray data were averaged across all samples from all donors in the matching anatomical region across both hemispheres. The data were also averaged across probes corresponding to the same gene, excluding probes that were not matched to gene symbols in the AIBS data. Two MRI regions were excluded as both the mean and the range of gene expression values in these regions were outliers compared to the other cortical regions of interest. The final output was a matrix of Z-scored expression values for each of 20,737 genes estimated in 306 MRI regions (see **SI** for detail). Gene expression data for individual genes such as Myelin Basic Protein was obtained from the appropriate row of this matrix. Similarly, gene expression values for larger gene sets of interest were extracted from the whole genome data. This included 94 genes that were identified as oligodendrocyte-specific by Cahoy et al (37) and 349 protein-coding genes recently identified as schizophrenia risk-genes (38), of which 312 were successfully matched to genes in the AIBS dataset.

Structural covariance and network analyses

The structural covariance matrix comprised the pair-wise correlations of cortical thickness for all possible pairs of regions in the parcellation template. Binary graphs were constructed to be node-connected with a connection density of 10% (39). From this graphical model of the connectome, we estimated two measures of topological centrality at each node: degree and closeness. These are representative of the more general concept of nodal centrality and they are strongly correlated. We also estimated the connection distance as the mean of the Euclidean distance of all the non-zero edges at each node. On average over all nodes, we estimated assortativity, clustering and global efficiency, and combined these measures of network segregation and integration, respectively, to estimate the small-worldness of the global network (59). The community structure of the graph was "nearly decomposed" by maximization of modularity (60) into a set of sparsely inter-connected modules or sub-graphs. The core/periphery structure of the graph was defined as a small rich club or core of highly inter-connected hubs, embedded in a larger set of more peripheral nodes. See **SI** for detail.

Statistical analyses

At each regional node, we fitted a simple linear regression model across all participants to estimate the gradient (ΔCT, ΔMT) and intercept or baseline (CT14, MT14) for each MRI measurement. For MT, the same model was fitted separately at each depth level between the pial surface and 0.8 mm below the GM/WM boundary. The null hypothesis of zero age-related change in ΔCT or ΔMT was tested globally on average over all regions and at each region individually, controlling the false discovery rate (FDR) at 5% to correct for multiple comparisons entailed by regional analysis. PLS regression on the gene expression matrix was used to identify the linear combinations of genes that best predicted the response variables (CT14, MT14, ΔCT, ΔMT). The statistical significance of the goodness-of-fit of the first two PLS components was tested with two-tailed $\alpha = 0.05$ by 1000 permutations of the response variables. The error in estimation of the weight of each gene on each PLS components was assessed by bootstrapping and the ratio of the weight of each gene to its bootstrap standard error was used to rank the genes according to their contribution to each PLS component. We used Gene Ontology (GO) enrichment analysis tools to identify and summarise annotations corresponding to biological processes that were significantly over-represented (FDR = 5%); see **SI** for detail. We predicted hypothetically that the transcriptional profile strongly associated with adolescent cortical

consolidation (PLS2) should be enriched for genes associated with oligodendroglia (37), or risk for schizophrenia (38). We tested for significant enrichment by a permutation test of the normalised bootstrap weight of each gene in PLS1 and PLS2, summed over all genes in the set.

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Figure Legends

Fig. 1: Cortical thickness (CT) and magnetisation transfer (MT) maps. At 14 years, primary somatosensory and visual cortex had thinner cortex (**a**) and greater MT (**c**) than association cortical areas. Baseline CT (mm) and MT (PU) were negatively correlated, confirming that thinner cortex was more myelinated at 14 years (**f**). Globally, over all 308 regions, CT decreased linearly with increasing age in the range 14 to 24 years (**b**) although there were regional differences in the rate of cortical shrinkage (ΔCT), with significantly non-zero rates of shrinkage (permutation test, FDR = 0.05) located mainly in association cortex (**e**). Globally, MT increased linearly with increasing age in the range 14 to 24 years (**d**) although there were regional differences in the rate of cortical myelination (ΔMT), with significantly non-zero rates of myelination (permutation test, FDR = 0.05) located mainly in association cortex (**g**). Rates of change in thickness and MT were negatively correlated confirming that more rapidly shrinking areas of cortex had faster rates of myelination (**h**).

Fig. 2: Magnetisation transfer mapping of intra-cortical myelination. The schematic (**a**) highlights estimation of MT on a contour (orange line) located at 70% of the cortical depth from the pial surface; and on another contour (green line) located 1 mm below the boundary between grey matter and white matter (yellow line). These and other distances can be located approximately in the context of cytoarchitectonic lamina of human neocortex (**c**). Baseline MT increases monotonically with distance from the pial surface (**b**) but the age-related increase in MT (ΔMT) was greatest at 70% cortical depth (**d**). The correlation between baseline MT and myelin basic protein (*MBP*) gene expression was also strongest at 70% cortical depth (**e**). Boxes represent the median and interquartile range over all 308 regions at each depth.

Fig. 3: Distinct gene expression profiles were specifically associated with cortical thickness and magnetisation transfer at 14 years, or with adolescent processes of cortical consolidation. The first partial least squares component (PLS1) identified a profile of genes that were over-expressed in occipital and somatosensory cortex (**a**), negatively correlated with baseline CT (**b**), and positively correlated with baseline MT (**c**). The second PLS component (PLS2) identified a profile of genes that were over-expressed in prefrontal cortex (**d**), negatively correlated with adolescent cortical shrinkage (ΔCT; **e**), and positively correlated with adolescent intra-cortical myelination (ΔMT; **f**). PLS2 was enriched for genes functionally related to synaptic transmission, glutamatergic signaling, and potassium ion channels, colour-coded by P value for significant enrichment, in the gene ontology of biological processes (**g**).

Fig. 4: Hubs of the connectome were located in regions with faster rates of cortical consolidation and were associated with the same gene expression profile as consolidation. The structural covariance network comprised hubs with high degree centrality (**a**) and high closeness centrality (**b**) that were concentrated anatomically in association cortical areas; in both panels a and b nodes are colored and sized by the corresponding centrality metric and square nodes represent members of the rich club. Both degree (red) and closeness centrality (green) were negatively correlated with cortical shrinkage (ΔCT; **c**), and positively correlated with adolescent increase in myelination (ΔMT; **d**). The gene expression profile associated with adolescent cortical consolidation (PLS2 from **Fig. 3**) was also significantly associated with degree and closeness centrality (**e**).