Methods

Study samples

The ENIGMA-schizophrenia DTI working group currently comprises 30 cohorts from 14 countries totalling 2,391 healthy controls and 1,984 individuals with schizophrenia (see **Figure 1**).

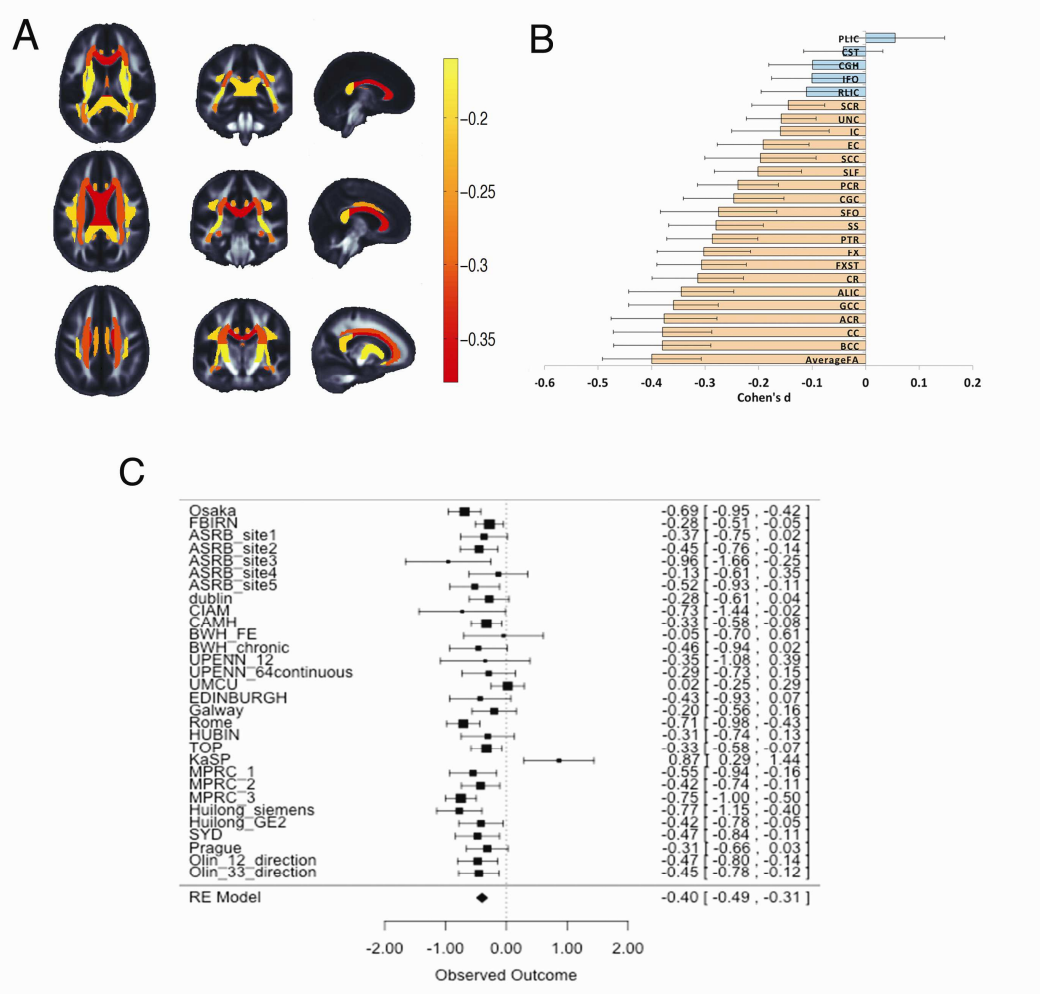

Figure 1. A. Comparison of FA between schizophrenia patients and healthy controls for 25 WM regions representing major fasciculi. B. The color bar indicates Cohen's *d* effect sizes after meta-analysis and meta analysis results, sorted in increasing magnitude of Cohen's *d* effect sizes across 30 cohorts for FA differences in schizophrenia patients (N=1,984) versus healthy controls (N=2,391), after including age, sex, age×sex, age^2 and age^2×sex, as covariates. Error bars represent 95% confidence intervals. Significant regions after adjusting for multiple regions tested ($p < 0.05/25 = 0.002$) are highlighted in orange. C. Forest plot of effect sizes for 30 cohorts. Interactive 3D visualization of the

The mean age across samples was 35.85 years for controls (range: 18-86) and 36.03 years for patients (range: 18-77). Samples of controls and patients were approximately 53.4% and 67% male respectively, but effects of sex were also modelled. The mean age at onset and duration of illness across the patient

groups were 23 and 15 years respectively. The mean total Positive and Negative Syndrome Scale (PANSS) and Scale for the Assessment of Positive and Negative Symptoms (SANS and SAPS) across the samples were 55.59,16.72 and 13.70 respectively. For samples that recorded current antipsychotic type and dose, the fraction of patients on second-generation antipsychotics (atypical) was 71%, first generation (typical) was 6%, both 10% and neither 13%. As in van Erp et al (2015), chlorpromazine (CPZ) equivalents were computed using methods previously described in Woods (2005;http://www.scottwilliamwoods.com/files/Equivtext.doc). The mean CPZ dose equivalent across the samples was 380.08. **Supplementary Tables 2-3** summarize key clinical and demographic information. Each study sample had been assessed with participants' written informed consent approved by local Institutional Review Boards.

Imaging acquisition and processing

Details of study type, scanner and acquisition parameters for each of the 30 sites are provided in **Supplementary Table 1**. Preprocessing, including eddy current correction, EPI induced distortion correction, and tensor fitting, was carried out at each site. Recommended protocols and procedures as well as quality control pipelines are available as part of the ENIGMA-DTI webpage and NITRC, but harmonization of preprocessing schemes was not enforced across sites to allow individual sites to use existing pipelines that may be more appropriate for their data acquisition. Once tensors were estimated and DTI measures of FA, harmonized image analysis was conducted at each site using the ENIGMA-DTI protocol (see Supplementary Note 1). Available mean diffusivity (MD), axial diffusivity (AD) and radial diffusivity (RD) images for a subset of samples were also derived, The present analysis combined ROIs across both hemispheres to avoid any potential issues of left/right flipping. Lateralized results are reported in **Supplementary table 4** but should be interpreted with caution.

Statistical analysis

Per-site analysis

We evaluated FA, and available MD,AD and RD differences between schizophrenia cases and healthy controls by calculating Cohen's *d* effect size estimates for diagnosis of schizophrenia in each of the 25 ROIs listed in **Table 1**.

To tease apart regional WM effects from global differences, post-hoc analyses on a sub-sample of available data from 17 cohorts (1,361 healthy controls and 1,226 patients) were carried out to covary for the effects of global FA measures across the entire skeleton, including average FA across the full skeleton, the central, or "core" FA comprised of the average of all voxels in the JHU ROIs, and the peripheral regions defined as everything besides the core in the full skeleton analysed (See Supplementary Note 2 for calculations of core and periphery FA).

Cohen's *d* effect sizes were also calculated for differences in FA between patients on atypical antipsychotics, typical antipsychotics, both, and unmedicated. Differences in FA were also assessed between subgroups of patients and controls who were smokers versus nonsmokers. Multiple linear regressions were performed to examine the effects of age at onset, duration of illness, CPZ scores, PANSS total, positive and negative scores, SAPS and SANS total scores. Age, sex, age-by-sex interaction, and quadratic covariates of age^2 and age^{2}-by-sex interaction were modeled as linear and nonlinear age and sex interactions have been reported for FA[22]. As age and duration of illness are collinear, we also examined duration of illness in years without covarying for age, as well as duration expressed as the percent of a person's lifetime they had been ill. Interaction effects, including diagnosis-by-sex and diagnosis-by-age were also calculated. A minimum of 10 subjects per group was used as the cut-off for inclusion in the statistical analyses.

All analyses were conducted using generalizable scripts available on the ENIGMA-GitHub https://github.com/ENIGMA-git/ENIGMA/tree/master/WorkingGroups/EffectSize_and_GLM. Individual sites download a single set of R scripts and specify the set of regressions that were customized for the current ENIGMA-Schizophrenia DTI analysis, publicly available on a set of Google Spreadsheet configuration files. Standardized regression outputs were then uploaded to a central server for meta analysis.

Meta-analysis

As in prior ENIGMA disease working group meta-analyses[21,23,24,25], a random-effects inverse-variance weighted meta-analysis was conducted at a central coordinating site (the University of Southern California Imaging Genetics Center) in R (metaphor package, version 1.99-118) to combine individual site effect sizes (See Supplementary Note 3). Heterogeneity scores (I^2) for each test were also computed, indicating the percent of the total variance in effect size explained by heterogeneity of the effects alone. Lower values of I^2 indicate lower variance in the effect size estimates across studies.

Effect sizes are reported as overall Cohen's *d* values for case/control effects and Z-scores for quantitative effects from linear regressions. To control the reporting of false positives from multiple tests across the ROIs, for our primary analysis of FA differences in cases as compared to controls, effects were declared to be significant if they survived the Bonferroni correction threshold of 0.05/25 = 0.002.

Results

FA differences between schizophrenia patients and controls

20 of 25 regions showed significantly lower FA in patients. Based on previous meta-analysis[5] we hypothesized that WM tracts interconnecting the frontal lobe, thalamus, cingulate gyrus and regions of the temporal lobe would be most severely affected. The largest effect size was observed for lower *average* FA (across the whole-brain WM skeleton) in schizophrenia patients, followed by body of the corpus callosum, the whole CC, the anterior *corona radiate* (ACR), and the *genu* of the CC (GCC). Significant patient reductions were also found in 21 other regions of interest (see **Figure 1 and Table 1**).

ROI	Cohen's d	SE	p-value	i^2	N voxels	N cases/controls
Average Skeletal FA	0.40	0.05	2.23×10^{-17}	48.14	112889	1984/2391
Core FA	0.41	0.05	9.70×10^{-16}	27.79	31742	1361/1226
Peripheral FA	0.47	0.05	3.75×10^{-22}	23.24	81147	1361/1226

BCC - body of *corpus callosum*	0.38	0.05	2.50×10^{-16}	46.57	3173	1984/2391
CC - *corpus callosum*	0.38	0.05	7.08×10^{-16}	48.03	7318	1984/2391
ACR - anterior *corona radiata*	0.38	0.05	7.88×10^{-14}	54.59	3129	1984/2391
GCC - *genu* of *corpus callosum*	0.36	0.04	4.34×10^{-17}	37.74	1834	1984/2391
ALIC - anterior limb of internal capsule	0.34	0.05	5.83×10^{-12}	54.13	1510	1984/2391
CR - corona radiata	0.31	0.04	5.72×10^{-13}	40.01	7344	1984/2391
FXST - fornix *stria terminalis*	0.31	0.04	7.06×10^{-13}	37.92	706	1984/2391
FX - fornix	0.30	0.04	1.16×10^{-11}	42.79	222	1984/2391
PTR - posterior thalamic radiation	0.29	0.04	3.87×10^{-11}	39.57	1987	1984/2391
SS - sagittal stratum	0.28	0.05	6.19×10^{-10}	44.32	1294	1984/2391
SFO - superior fronto-occipital fasciculus	0.27	0.06	7.35×10^{-7}	62.84	193	1984/2391
CGC - cingulum (cingulate gyrus)	0.25	0.05	2.61×10^{-7}	50.22	594	1984/2391
PCR - posterior corona radiata	0.24	0.04	6.50×10^{-10}	25.58	1437	1984/2391
SLF - superior longitudinal fasciculus	0.20	0.04	1.30×10^{-6}	35.17	3503	1984/2391
SCC - superior corona radiata	0.20	0.05	0.00021	59.30	2311	1984/2391
EC - external capsule	0.19	0.04	1.17×10^{-5}	40.88	2896	1984/2391
IC - internal capsule	0.16	0.05	0.0006	47.33	4781	1984/2391
UNC - uncinate	0.16	0.03	2.11×10^{-6}	5.02	125	1984/2391
SCR - superior corona radiata	0.14	0.03	3.11×10^{-5}	11.24	2778	1984/2391
RLIC - retrolenticular part of IC	0.11	0.04	0.011	40.25	1496	1984/2391
IFO - inferior fronto-occipital fasciculus	0.10	0.04	0.009	26.03	88	1984/2391
CGH - cingulum (hippocampal portion)	0.10	0.04	0.018	35.96	524	1984/2391
CST - corticospinal tract	0.04	0.04	0.267	23.14	167	1984/2391
PLIC - posterior limb of IC	0.05	0.05	0.245	49.48	1775	1984/2391

Table 1. Cohen's *d* values, their standard errors (SE), *p*-values and I2 (heterogeneity) values after meta-analysis, for FA differences between schizophrenia patients and healthy controls.