# Untangling the adolescent internalizing brain: investigations on brain networks in youth with anxious and depressive problems

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# 5

# White matter microstructure alterations in social anxiety disorder – a mega-analysis across 12 cohorts in the ENIGMA-Anxiety Working Group

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## Abstract

**Background:** Studies investigating social anxiety disorder (SAD) have reported inconsistent alterations in white matter (WM) microstructure. The ENIGMA-Anxiety Working Group investigated differences in microstructure of 25 WM tracts between individuals with SAD and healthy controls in a mega-analysis.

**Methods:** We analyzed data from 487 individuals with SAD and 1,604 healthy controls (HC) (age 8 – 65) from twelve cohorts worldwide. Analyses and quality control were performed using standardized ENIGMA diffusion tensor imaging (DTI)-protocols. We primarily examined fractional anisotropy (FA) as the main parameter of WM microstructure. Linear mixed-effects analyses were conducted to compare individuals with SAD with HC in the full sample. Next, adult (age > 21) and adolescent (age ≤ 21) samples were analyzed separately. In sensitivity analyses, additional effects of sex, medication, symptom severity and comorbid psychiatric disorders were investigated.

**Results:** In the full sample, individuals with SAD showed lower FA in several tracts, including the corpus callosum and fornix, when compared to HC. Widespread sex-by-diagnosis interactions were observed, mostly driven by lower FA in SAD females. Adults with SAD showed lower FA in multiple tracts, while age-by-diagnosis interactions were observed in adolescents.

**Conclusions:** Using a mega-analytic approach, several differences in WM microstructure were found between individuals with SAD and HC, both in the full sample and in age-group specific sensitivity analyses. Some neurobiological changes in WM tracts in individuals with SAD may vary with age and sex, whereas others might relate to broader transdiagnostic neurobiological features underlying psychopathology. Further research should investigate these issues in more detail.

## Introduction

Social anxiety disorder (SAD) is one of the most prevalent and debilitating psychiatric disorders with a lifetime prevalence rate between 4 – 13 % and a typical onset in childhood or early adolescence, often with high psychiatric comorbidity and a chronic course [1, 2]. The pathogenesis of SAD involves complex interactions among biological, environmental, genetic, and temperamental factors [2, 3]. Neurobiological factors may help to inform current treatments and identify possible targets for new interventions.

Various modalities of magnetic resonance imaging (MRI) uncovered altered brain structure, functioning and connectivity in individuals with SAD [4, 5]. A review of prior work examined white matter (WM) microstructure assessed with diffusion tensor imaging (DTI) in small groups of individuals with a primary diagnosis of SAD compared to healthy controls (HC) [6]. These DTI studies revealed that WM microstructure differed from HC in several regions, mostly using fractional anisotropy (FA) as a general measure of WM microstructure. Moreover, lower FA of the uncinate fasciculus (UF) in SAD compared to HC has been reported in some studies using whole brain voxel-wise analysis or fiber tractography [7-9], although this finding has not been replicated in other studies [10-13]. Additionally, some studies reported negative correlations between anxiety symptoms and FA in the right inferior fasciculus (ILF) and superior longitudinal fasciculus (SLF) in individuals with SAD [10, 12, 13]. Taken together, there is little consistency between affected WM regions reported, and most of these studies had low sample sizes (ranging from $n$ = 36 to $n$ = 88 for the total sample).

Within the framework of the Enhancing NeuroImaging Genetics through Meta Analysis (ENIGMA)-Anxiety Working Group [14], we initiated a worldwide multi-site analysis to investigate WM microstructure in SAD. The present study consisted of a mega-analysis of harmonized pooled individual participant data (IPD) [15], including 487 individuals with SAD and 1,604 HC from twelve samples worldwide. Our main objective was to explore differences in WM microstructure between individuals with SAD and HC in major tracts of the brain. In addition, we explored age-specific SAD-related alterations in WM microstructure by conducting sensitivity analyses in adults and adolescents separately, as WM maturation is nonlinear and heterochronicity in this maturation has been associated with psychiatric disorders [16, 17]. Furthermore, we investigated SAD-related WM interactions with age and sex, WM differences in relation to psychiatric comorbidity, medication use and symptom severity. Lastly, we investigated whether WM differences in SAD correlated with WM differences in other psychiatric disorders. As these analyses were conducted in the largest dataset on WM microstructure in SAD to date, we aim to reduce the chance of false positives and thus to increase the accuracy of our findings [18].

## Methods and materials

### Participants

Individual-level participant data (IPD) from previous neuroimaging studies on SAD were considered for inclusion if DTI-data were collected during MRI-scanning [15]. Cohorts were included if participants had a current or lifetime diagnosis of SAD (criterion 1), also consisted of a group of HC without any lifetime psychiatric disorder (criterion 2) and had at least collected information on sex and age for all participants (criterion 3). For the patient group, current or lifetime diagnoses of comorbid major depressive disorder (MDD), generalized anxiety disorder (GAD), panic disorder, agoraphobia, specific phobia, obsessive-compulsive disorder (OCD), post-traumatic stress disorder (PTSD), and substance use dependence were allowed. However, individuals with SAD were excluded if they had a current diagnosis or history of psychosis, schizophrenia, bipolar disorder or autism spectrum disorder of at least moderate severity. The diagnostic assessment measures and exclusion criteria for every cohort can be found in Table S1.

Local institutional review boards and ethics committees approved the individual research protocols. Written informed consent was obtained for all adult participants and parents of participants younger than 18 years old at the local research sites. In addition, principal investigators from each site signed a memorandum of understanding. This memorandum included guidelines on subject de-identification, data sharing, and confidentiality and security practices [14]. Furthermore, approval from local officials to share data was obtained by principal investigators from each site, or via research contract offices when raw data was to be shared.

Ten ENIGMA-Anxiety Working Group sites shared their IPD, after locally processing their data using the ENIGMA-DTI processing pipeline (http://enigma.ini.usc.edu/protocols/dti-protocols/), while for the Muenster cohort, raw imaging data was shared and processed subsequently at the coordinating site (Leiden). In addition, raw data from the publicly available Philadelphia Neurodevelopmental Cohort was downloaded and processed in Leiden.

Total sample size of IPD after processing and quality control consisted of 487 individuals with SAD and 1,604 HC. More specifically, the adult sample (age 22 – 65 years) contained data from 230 individuals with SAD and 1301 HC; the adolescent sample (age 8 – 21 years) included 257 individuals with SAD and 303 HC. Demographic information and clinical characteristics for every cohort are summarized in Table 1, Table 2 and Table S2.

### Image (pre)-processing

Information on scanner and acquisition parameters is presented in Table S3. At each site, preprocessing of diffusion weighted images was performed, which included eddy current correction, echo-planar imaging (EPI) induced distortion correction and tensor fitting. After preprocessing, data were processed according to the ENIGMA-DTI protocol (available at https://enigma.ini.usc.edu/protocols/dti-protocols/) using the FMRIB Software Library (FSL). Tract-based spatial statistics (TBSS) were applied

as follows. First, individual tensor images were analyzed, resulting in individual level images of FA and additional measures of WM microstructure, being mean diffusivity (MD), radial diffusivity (RD) and axial diffusivity (AD). Then, quality control procedures were conducted, including visual inspection of FA and registration and projection onto the standardized ENIGMA skeleton. Finally, individual mean values of FA, MD, AD and RD were obtained for 25 regions of interest (ROI), covering most WM regions in the brain. These IPD-results from the local sites were sent to the coordinating site in Leiden. Here, final quality control measures were performed following the ENIGMA-DTI protocol, including inspection of histograms of each WM parameter per ROI per cohort to assess normality and FA outliers (> 5 SD), using an automated protocol. No outliers were detected. Afterwards, results were merged into one dataset. As no lateralized effects were hypothesized, bilateral averaged measures were used in the primary analysis, by combining all ROIs across both hemispheres (mean of the left and right hemisphere regions weighted by the number of voxels). Lastly, we optimized harmonization of FA by applying eHarmonize, a newly developed python package, to match site-specific FA to lifespan reference curves [19].

### Regions of interest

In total, 25 ROIs were investigated. We examined the average FA of 24 WM ROIs, of which 21 are illustrated in Figure 1, and the average FA over all WM regions. Three additional regions not illustrated in Figure 1 comprise several subregions described in the figure, being 1) the corpus callosum (CC), consisting of the body (BCC), genu (GCC) and splenium of the corpus callosum (SCC); 2) corona radiata (CR), consisting of an anterior (ACR), posterior (PCR) and superior (SCR) part; and 3) the internal capsule (IC), consisting of the anterior limb (ALIC), posterior limb (PLIC) and the retrolenticular limb (RLIC). The inferior longitudinal fasciculus (ILF) was included as part of the sagittal striatum (SS), in line with the ENIGMA-DTI protocol.

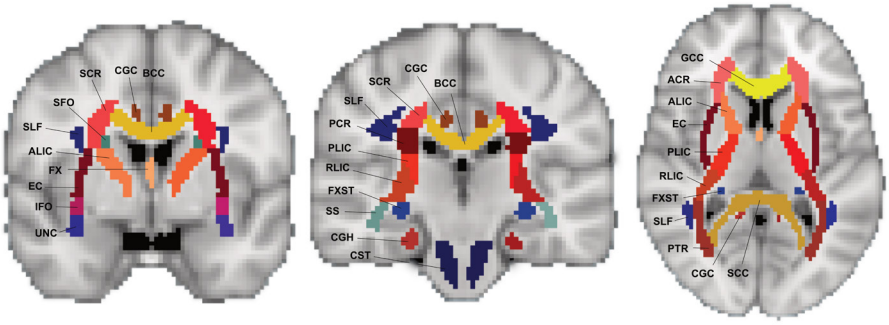



**Figure 1 Illustration of white matter ROIs used to investigate differences in microstructure between individuals with SAD and HC participants.** *Abbreviations:* ACR: anterior corona radiata (left and right); ALIC: anterior limb of internal capsule (left and right); BCC: body of corpus callosum; CGC: cingulum (cingulate gyrus; left and right); CGH: cingulum (hippocampal portion; left and right); CST: corticospinal tract (left and right); EC: external capsule (left and right); FX: fornix; FXST: fornix (crues) / stria terminalis (left and right); GCC: genu of corpus callosum; IFO: inferior fronto-occipital fasciculus (left and right); PCR: posterior corona radiata (left and right); PLIC: posterior limb of internal capsule (left and right); PTR: posterior thalamic radiation (left and right); RLIC: retrolenticular part of internal capsule (left and right); SCC: splenium of corpus callosum; SCR: superior corona radiata (left and right); SFO: superior fronto-occipital fasciculus (left and right); SLF: superior longitudinal fasciculus (left and right); SS: sagittal stratum (left and right); UNC: uncinate fasciculus (left and right).

**Linear mixed-effects models**

This study was preregistered in December 2023 at https://osf.io/5ycag/. In light of additional insights gained during the analysis process, minor refinements were made to the analytic approach (Note S1). Using linear mixed-effects models (cf. previous work from the ENIGMA-Anxiety Working Group in SAD) [20], we investigated FA for each of the 25 WM ROIs as primary outcome variable, with SAD diagnosis (dichotomous factor) or symptom severity (continuous factor) as main regressor. We used age, sex, age$^2$, age-by-sex and age$^2$-by-sex as covariates. Age was centred throughout. Mixed-effects *d* effect sizes were calculated from the t-values for diagnostic factor, and mixed-effects *r* estimates were calculated for relevant interaction and continuous variables of interest. These are similarly scaled as Cohen's *d* estimates and *r* estimates, but include a correction for non-independence in the aggregated dataset [21]. For all analyses, samples were only included for between-group contrasts when at least one observation per group was available for each of the WM regions. The threshold for significance was set at a false-discovery rate (FDR) corrected $p < 0.05$, adjusting for 25 WM regions in all analyses, in line with previous ENIGMA-DTI projects [22]. Because development of WM regions is known to peak during childhood and adolescence [16], we explored FA within the full sample and in adults (> 21 years old) and adolescents (≤ 21 years old) separately. Additional WM parameters, being MD, AD and RD, were only investigated if a significant difference in FA was found.

First, we investigated if average FA in each of the 25 predefined WM ROIs was different between individuals with SAD and HC. Then, diagnosis-by-age, diagnosis-by-age$^2$ and diagnosis-by-sex interactions were investigated. Next, we conducted sensitivity analyses to investigate possible confounders. In these analyses, individuals with SAD were excluded if they had comorbid lifetime PTSD, OCD, or when current SAD criteria were not met. Furthermore, we compared subgroups of individuals with SAD to HC to investigate individuals with SAD with and without comorbid lifetime MDD, with and without any comorbid anxiety disorders and with and without psychotropic medication use at the time of scanning, specifically investigating individuals with SAD using selective serotonin reuptake inhibitors (SSRI) and serotonin-noradrenaline reuptake inhibitors (SNRI), as their pharmacological mechanisms overlap. In addition, we investigated associations between WM characteristics and clinical symptoms within the SAD group, using the Liebowitz Social Anxiety Scale (LSAS) to assess severity of social anxiety, the Beck Depression Inventory II (BDI-II) to assess severity of depressive symptoms and the State-Trait Anxiety Inventory (STAI) for symptoms of trait anxiety, when available [23-25].

Lastly, as symptoms of social anxiety are common comorbidities across many psychiatric illnesses [26, 27], we performed correlation analyses between mega-analytical effect sizes observed in this study and the regional patterns of patient-control deficits derived using the same ENIGMA-DTI workflow for five other psychiatric illnesses, distributed as part of ENIGMA RVI package 28. These illnesses include schizophrenia, bipolar disorder, MDD, PTSD and OCD; findings of these studies are published before [22, 28-31].

### Selection of mega analytic approach

We selected the model for our mega-analytic approach as previously reported by Groenewold and colleagues [20]. All linear mixed-effects models were fitted with a random-intercept to account for data clustering within samples. Next, models with a random slope for diagnosis per sample (complex model) and without random slope (reference model) were fitted, and fit was compared using the Likelihood Ratio Test (LRT; cf. Boedhoe and colleagues) [32], where $p < 0.05$ indicates improved model fit in complex relative to reference model. The mega-analytic model with the best model fit for most WM regions was selected, based on the full sample. All mega-analysis models were fitted with restricted maximum likelihood (ReML) in R version 4.3.0 and mixed-effects *d* and *r* effect sizes were computed (Table S4) [33].

## Results

### Participants

Demographic and clinical characteristics for each cohort are presented in Table 1 and Table 2.

**Table 1 Demographic information for samples included in current mega-analyses.**

| | Cohort | Country | Full sample | | | | | | Adult samples (age > 21) | | | | | | Adolescent samples (age ≤ 21) | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | Total no. | | Age (mean ± SD) | | % Female | | Total no. | | Age (mean ± SD) | | % Female | | Total no. | | Age (mean ± SD) | | % Female | |
| | | | HC | SAD | HC | SAD | HC | SAD | HC | SAD | HC | SAD | HC | SAD | HC | SAD | HC | SAD | HC | SAD |
| 1 | Istanbul | TR | 22 | 22 | 28.7 ± 6.6 | 28.7 ± 6.6 | 50.0 | 50.0 | 22 | 20 | 28.7 ± 6.6 | 29.7 ± 6.2 | 50.0 | 45.0 | | 2 | | 19.5 ± 0.7 | | 100.0 |
| 2 | LFLSAD | NL | 10 | 10 | 47.2 ± 12.9 | 45.3 ± 5.3 | 30.0 | 70.0 | 9 | 10 | 50.4 ± 8.3 | 45.3 ± 5.3 | 22.2 | 70.0 | 1 | | 17.9 ± NA | | 100.0 | |
| 3 | FOR2107-MR | DE | 511 | 31 | 36.6 ± 13.2 | 32.7 ± 10.2 | 62.8 | 74.2 | 476 | 27 | 37.9 ± 12.8 | 34.5 ± 9.7 | 61.8 | 77.8 | 35 | 4 | 19.7 ± 1.3 | 20.5 ± 0.6 | 77.1 | 50.0 |
| 4 | FOR2107-MS | DE | 342 | 57 | 31.3 ± 12.1 | 35.2 ± 12.7 | 67.5 | 70.2 | 298 | 49 | 33.0 ± 12.0 | 37.7 ± 12.1 | 66.1 | 67.3 | 44 | 8 | 19.9 ± 1.0 | 20.3 ± 0.9 | 77.3 | 87.5 |
| 5 | Muenster | DE | 24 | 22 | 25.9 ± 6.0 | 28.7 ± 8.3 | 62.5 | 54.5 | 22 | 21 | 26.6 ± 5.8 | 29.0 ± 8.3 | 59.1 | 52.4 | 2 | 1 | 18.5 ± 0.7 | 21.0 ± NA | 100.0 | 100.0 |
| 6 | MNC | DE | 466 | 58 | 37.0 ± 11.8 | 32.9 ± 11.1 | 56.4 | 79.3 | 429 | 54 | 38.5 ± 11.0 | 34.0 ± 10.8 | 56.2 | 79.6 | 37 | 4 | 19.7 ± 1.6 | 18.8 ± 1.7 | 59.5 | 75.0 |
| 7 | TIP | DE | 37 | 39 | 26.2 ± 9.3 | 25.1 ± 6.7 | 73.0 | 74.4 | 22 | 24 | 30.3 ± 10.1 | 28.3 ± 6.8 | 63.6 | 70.8 | 15 | 15 | 20.1 ± 0.9 | 19.9 ± 0.9 | 86.7 | 80.0 |
| 8 | NIMH-SDAN | US | 43 | 24 | 12.8 ± 2.6 | 11.9 ± 2.6 | 48.8 | 62.5 | | | | | | | 43 | 24 | 12.8 ± 2.6 | 11.9 ± 2.6 | 48.8 | 62.5 |
| 9 | PNC | US | 113 | 185 | 13.8 ± 3.5 | 15.5 ± 3.0 | 46.9 | 57.8 | 2 | 1 | 22.0 ± 0.0 | 22.0 ± NA | 50.0 | 100.0 | 111 | 184 | 13.6 ± 3.4 | 15.4 ± 2.9 | 46.8 | 57.6 |
| 10 | MRC-SU | SA | 14 | 16 | 30.4 ± 8.0 | 29.9 ± 9.4 | 50.0 | 68.8 | 11 | 14 | 33.2 ± 6.7 | 31.4 ± 9.2 | 54.5 | 64.3 | 3 | 2 | 20.3 ± 0.6 | 20.0 ± 1.4 | 33.3 | 100.0 |
| 11 | Vanderbilt | US | 5 | 6 | 23.0 ± 2.0 | 22.0 ± 1.5 | 60.0 | 50.0 | 3 | 3 | 24.3 ± 1.2 | 23.0 ± 1.7 | 66.7 | 66.7 | 2 | 3 | 21.0 ± 0.0 | 21.0 ± 0.0 | 50.0 | 33.3 |
| 12 | West China Hospital | CN | 17 | 17 | 21.9 ± 4.0 | 21.9 ± 4.0 | 29.4 | 29.4 | 7 | 7 | 25.4 ± 4.0 | 25.4 ± 4.0 | 14.3 | 14.3 | 10 | 10 | 19.4 ± 0.8 | 19.4 ± 0.8 | 40.0 | 40.0 |
| | Total across all samples | | 1604 | 487 | 32.7 ± 13.5 | 24.1 ± 6.5 | 59.9 | 63.4 | 1301 | 230 | 36.4 ± 5.7 | 33.3 ± 9.2 | 60.1 | 67.0 | 303 | 257 | 16.5 ± 4.0 | 16.0 ± 2.5 | 58.4 | 60.3 |

*Abbreviations:* HC: healthy control; SAD: social anxiety disorder; SD: standard deviation; TR: Turkey; NL: Netherlands; DE: Germany; US: United States of America; SA: South-Africa; CN: China; NA: not applicable.
*Notes:* Sites were excluded from analysis if $n = 0$ for either diagnostic group.

**Table 2 Clinical characteristics for individuals with SAD in samples included in mega-analyses.**

| | Cohort | Full sample | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | Lifetime comorbidity | | Psychotropic medication | | LSAS | STAI-T | BDI-II |
| | | MDD (%) | ANX (%) | % any use | % SSRI / SNRI | mean ± SD | mean ± SD | mean ± SD |
| 1 | Istanbul | 0.0 | 0.0 | 0.0 | - | 73.9 ± 28.5 | - | - |
| 2 | LFLSAD | 50.0 | 40.0 | 10.0 | 100.0 | 60.6 ± 25.6 | 42.7 ± 9.2 | 13.5 ± 10.4 |
| 3 | FOR2107-MR | 98.2 | 28.1 | 64.9 | 89.2 | 0.0 ± 0.0 | 60.2 ± 10.3 | 21.7 ± 10.3 |
| 4 | FOR2107-MS | 96.8 | 41.9 | 83.9 | 80.8 | 0.0 ± 0.0 | 57.8 ± 8.9 | 20.7 ± 10.3 |
| 5 | Muenster | - | - | - | - | - | - | - |
| 6 | MNC | 46.6 | 12.1 | 24.1 | 100.0 | 60.8 ± 19.7 | 55.3 ± 11.6 | 16.6 ± 11.1 |
| 7 | TIP | 82.1 | 0.0 | 28.2 | 100.0 | 65.2 ± 21.6 | 51.6 ± 10.3 | 13.8 ± 10.4 |
| 8 | NIMH-SDAN | 0.0 | 83.3 | 0.0 | - | - | 42.2 ± 6.8 | - |
| 9 | PNC | 15.1 | 42.6 | 58.9 | 0.0 | - | - | - |
| 10 | MRC-SU | 50.0 | 0.0 | 0.0 | - | 88.3 ± 28.5 | - | - |
| 11 | Vanderbilt | 0.0 | 66.7 | 0.0 | - | | 25.7 ± 5.8 | 6.3 ± 4.6 |
| 12 | West China Hospital | 0.0 | 0.0 | 0.0 | - | 50.2 ± 12.8 | 0.0 ± 0.0 | 0.0 ± 0.0 |
| | *Total across all samples* | 39.9 | 30.9 | 44.7 | 40.4 | 65.1 ± 21.9 | 21.9 ± 53.2 | 53.2 ± 9.7 |

*Abbreviations:* -: information not available for sample; LSAS: Liebowitz Social Anxiety Scale, total score; STAI-T: State-Trait Anxiety Inventory – Trait subsection; BDI-II: Beck Depression Inventory, 2nd edition; ANX: any comorbid anxiety disorder; MDD: major depressive disorder; SSRI: selective serotonin reuptake inhibitor; SNRI serotonin-norepinephrine reuptake inhibitor.

**Mega-analytic results for comparisons in SAD vs HC**

Model fit comparisons were conducted on the full dataset ($n$ = 487 SAD, $n$ = 1,604 HC). For all ROIs and average FA over the whole WM skeleton, the complex model with random intercept (scan site) and random slope (SAD diagnosis per scan site) did not show a significant improvement in model fit when compared to the random intercept (scan site) reference model (Table S4). Thus, all subsequent analyses were conducted with the random intercept (scan site) model.

**Mega-analytic results for comparisons of SAD vs HC and relations with comorbidity in SAD subgroups**

To enhance readability, significant results are summarized in Table 3 and described below, where we report our findings by clustering significant regions displaying similar patterns. Additionally, Figure 2 displays results from the main analysis in the full sample as well as adolescent and adult samples separately. For a systematic and comprehensive overview of all analyses, we refer to Note S2 and Tables S5- S34.

In our main analyses of the full sample, lower FA was found in several regions. First, individuals with SAD had lower average FA when compared to HC, as well as lower FA in the FX, BCC and CC (Figure 2B). This pattern remained throughout several sensitivity analyses excluding individuals with SAD and comorbid PTSD (Table S8), OCD (Table S7) or other anxiety disorders (Table S12) and when including only individuals with SAD who had a current SAD diagnosis (Table S6; Note S2). We found a largely similar pattern in the adult only sample (Table S9) and in sensitivity analyses in adult individuals with SAD with (Table S17) and without comorbid depression (Table S18) and without comorbid anxiety disorders (Table S16). Moreover, findings in the CC and BCC were coupled with higher RD in the adult sample (Table 3).

Furthermore, individuals with SAD had lower FA in the SFO when compared to HC, an association also seen in adult individuals with SAD (Table S9) and in SAD patients with comorbid anxiety disorders (Table S11). Lastly, lower FA was found in the CR (Figure 2B). This finding seemed to be driven by adult individuals with SAD (Table S9).

Separate analyses on the adult sample revealed additional ROIs displaying SAD-related alterations in FA, namely lower FA in GCC, CGC and PTR (Table S9), even though we did not find significant age-by-diagnosis interactions in these regions in the full sample.

Analyses in the adolescent samples separately did not reveal significant differences in any of the ROIs when correcting for multiple comparisons (Table S10). There were no significant results when investigating the role of psychotropic medication or associations with symptom severity in the full sample, adult or adolescent samples separately (Tables S23 – S34).

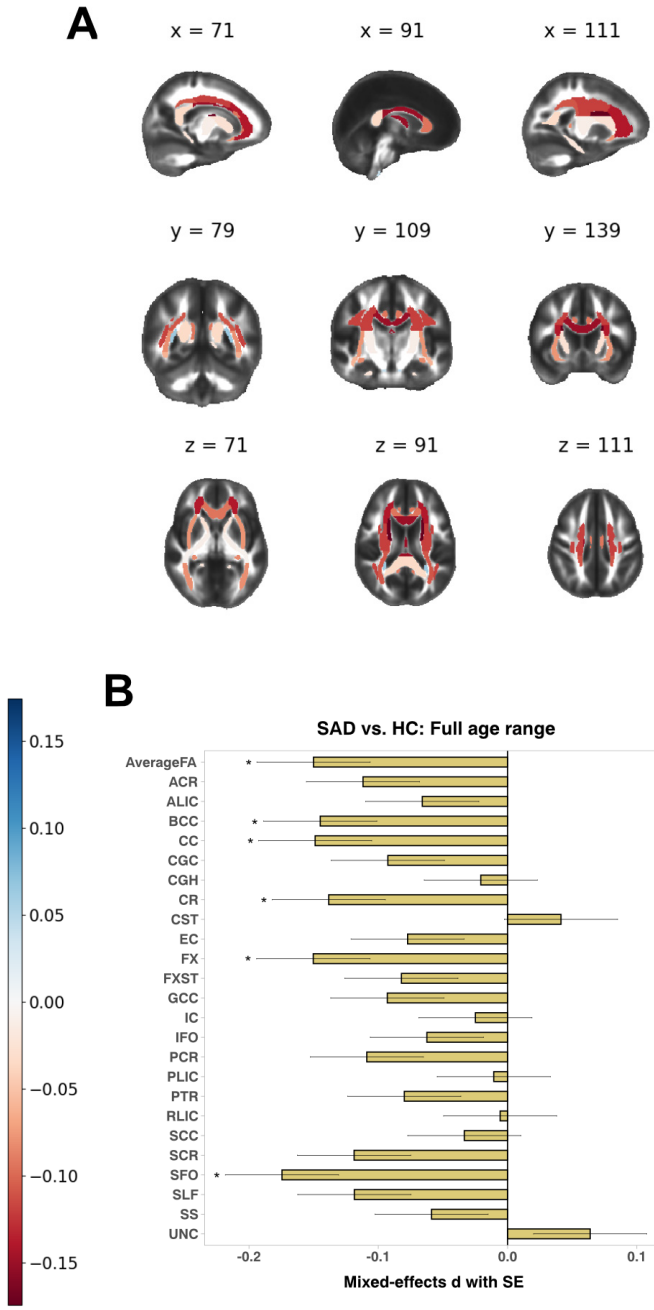



**Figure 2 Differences in white matter microstructure between individuals with social anxiety disorder (SAD) and healthy controls (HC) in 25 white matter tracts.**

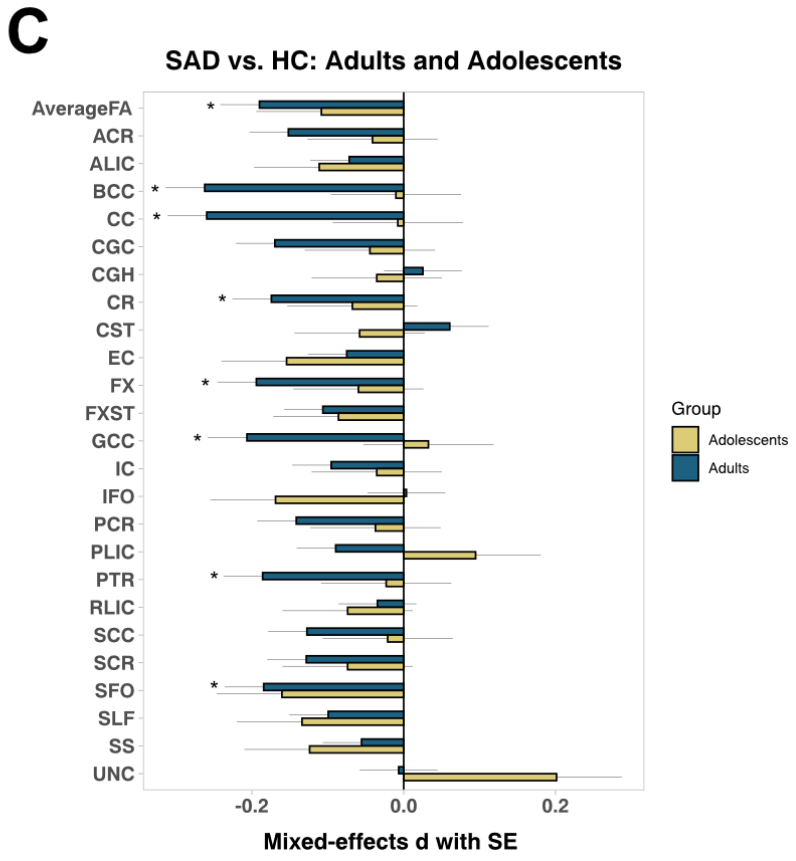



**Figure 2 (continued)**

*Notes:* A. Illustration of differences in FA between patients with SAD and healthy controls in 25 white matter regions. Color bar represents mixed-effects *d* effect size. Blue represents greater FA in SAD compared to HC. Red represents greater FA in HC compared to SAD. B. Mixed-effects *d* effect size in the full age range. Error bars represent standard error (SE). C. Mixed-effects *d* effect size stratified according to adult (> 21 years old) and adolescent age group (≤ 21 years old). Error bars represent standard error (SE). $^{*}p \leq 0.05$ after false-discovery rate adjustment for multiple comparisons.

**Table 3 Overview of mega-analytic results for main diagnostic group comparisons and clinical subgroup comparisons.**

| Group comparison | Sens | SAD | HC | averageFA[a] | | BCC | | CC | | GCC | | CR | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Age | n | n | mixedeff.d | pFDR[bc] | mixedeff.d | pFDR | mixedeff.d | pFDR | mixedeff.d | pFDR | mixedeff.d | pFDR |
| SAD vs HC | - | 487 | 1605 | -0.15 | 0.026 | -0.15 | 0.026 | -0.15 | 0.026 | ns | ns | -0.14 | 0.032 |
| SAD vs HC | Adu | 230 | 1301 | -0.19 | 0.036 | -0.26[d] | 0.004 | -0.26[d] | 0.004 | -0.21 | 0.033 | -0.17 | 0.047 |
| SAD vs HC | Ado | 255 | 292 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| SAD ANX vs. HC | - | 142 | 1490 | -0.23 | 0.074 | ns | ns | ns | ns | ns | ns | -0.22 | 0.074 |
| SAD ANX vs. HC | Adu | 36 | 1217 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| SAD ANX vs. HC | Ado | 106 | 262 | -0.23 | 0.273 | ns | ns | ns | ns | ns | ns | ns | ns |
| SAD wo ANX vs. HC | - | 317 | 1559 | -0.14 | 0.129 | -0.20 | 0.032 | -0.19 | 0.032 | ns | ns | ns | ns |
| SAD wo ANX vs. HC | Adu | 170 | 1277 | -0.23 | 0.036 | -0.34[d,e] | 0.001 | -0.33[d] | 0.001 | -0.24 | 0.025 | -0.18 | 0.076 |
| SAD wo ANX vs. HC | Ado | 145 | 269 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| SAD MDD vs. HC | - | 184 | 1493 | ns | ns | -0.21 | 0.100 | -0.19 | 0.101 | ns | ns | -0.19 | 0.101 |
| SAD MDD vs. HC | Adu | 130 | 1245 | -0.19 | 0.144 | -0.30 | 0.032 | -0.25 | 0.046 | ns | ns | -0.22 | 0.087 |
| SAD MDD vs. HC | Ado | 54 | 232 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| SAD wo MDD vs. HC | - | 277 | 1403 | -0.16 | 0.167 | ns | ns | ns | ns | ns | ns | ns | ns |
| SAD wo MDD vs. HC | Adu | 76 | 1112 | -0.26 | 0.068 | -0.29 | 0.068 | -0.35 | 0.039 | -0.33 | 0.039 | ns | ns |
| SAD wo MDD vs. HC | Ado | 199 | 221 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |

**Table 3** (continued)

| Group comparison | Sens | SAD | HC | CGC | | FX | | PTR | | SFO | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | Age | n | n | mixedeff.d | pFDR | mixedeff.d | pFDR | mixedeff.d | pFDR | mixedeff.d | pFDR |
| SAD vs HC | - | 487 | 1605 | ns | ns | -0.15 | 0.026 | ns | ns | -0.17 | 0.019 |
| SAD vs HC | Adu | 230 | 1301 | -0.17 | 0.050 | -0.19 | 0.036 | -0.19 | 0.036 | -0.19 | 0.036 |
| SAD vs HC | Ado | 255 | 292 | ns | ns | ns | ns | ns | ns | ns | ns |
| SAD ANX vs. HC | - | 142 | 1490 | ns | ns | -0.21 | 0.088 | ns | ns | -0.28 | 0.034 |
| SAD ANX vs. HC | Adu | 36 | 1217 | ns | ns | ns | ns | ns | ns | -0.51 | 0.069 |
| SAD ANX vs. HC | Ado | 106 | 262 | ns | ns | ns | ns | ns | ns | ns | ns |
| SAD wo ANX vs. HC | - | 317 | 1559 | ns | ns | -0.14 | 0.129 | ns | ns | -0.16 | 0.077 |
| SAD wo ANX vs. HC | Adu | 170 | 1277 | -0.22 | 0.041 | -0.19 | 0.074 | -0.20 | 0.069 | ns | ns |
| SAD wo ANX vs. HC | Ado | 145 | 269 | ns | ns | ns | ns | ns | ns | ns | ns |
| SAD MDD vs. HC | - | 184 | 1493 | ns | ns | -0.24 | 0.069 | -0.17 | 0.101 | -0.16 | 0.124 |
| SAD MDD vs. HC | Adu | 130 | 1245 | ns | ns | -0.26 | 0.046 | -0.25 | 0.046 | ns | ns |
| SAD MDD vs. HC | Ado | 54 | 232 | ns | ns | ns | ns | ns | ns | ns | ns |
| SAD wo MDD vs. HC | - | 277 | 1403 | ns | ns | ns | ns | ns | ns | -0.18 | 0.150 |
| SAD wo MDD vs. HC | Adu | 76 | 1112 | -0.33 | 0.039 | ns | ns | ns | ns | -0.26 | 0.068 |
| SAD wo MDD vs. HC | Ado | 199 | 221 | ns | ns | ns | ns | ns | ns | ns | ns |

*Abbreviations:* Sens: sensitivity analysis by age group; SAD: social anxiety disorder; HC: healthy control; N: number of participants; BCC: body of corpus callosum; CGC: cingulum (cingulate gyrus); CR: corona radiata; FX: fornix; GCC: genu of corpus callosum; PTR: posterior thalamic radiation; SCR: superior corona radiata; SFO: superior fronto-occipital fasciculus; Mixeff.d: mixed-effects; FDR: false discovery rate correction; ns: non- significant (uncorrected $p < 0.05$); Adu: adults; Ado: adolescents; ANX: any comorbid anxiety disorder; wo: without. *Notes:* [a] Results presented for all regions with an FDR significant finding in one of the full age range mega-analyses. [b] Bold text highlights significant result after FDR multiple comparison correction. [c] Plain text indicates uncorrected significant result. [d] This decrease in FA was coupled with higher RD. [e] This decrease in FA was coupled with higher MD.

### SAD interactions with age

In the full sample, significant interaction effects between diagnosis and age were observed in FA for the CST and IFO (Table S35). Additional analyses revealed interaction effects for AD in the CST, but there were no significant interaction effects for MD or RD. There were no significant interactions between diagnosis and age in the adult sample (Table S36). When adolescent participants were considered, significant diagnosis-by-age interaction effects were observed for the CST, IFO and PTR (Table S37). Follow-up analyses highlighted significant effects for RD in IFO and PTR. No significant interaction effects were found for AD or MD. Illustrating post-hoc plots showed no clear consistent direction of these interactions (Figure S1). There were no significant diagnosis-by-age$^2$ interaction effects in the full, adult or adolescent samples (Tables S38-40).

### SAD interactions with sex

Significant diagnosis-by-sex interactions for FA were observed in 19 out of the 25 ROIs across the full sample (Table S41). The largest effects were found across the WM skeleton, SLF, CR, SCR, CST, FX, ALIC, IC, PCR, ACR and PTR. Post-hoc tests revealed that in most regions, effects were driven by lower FA in females with SAD, although in some regions, effects were driven by lower FA in males with SAD (Figure 3 and Figure S2). Other significant interactions, but with smaller effect sizes, were found in the CGC, SFO, CGH, RLIC, EC, PLIC, CC and GCC (Table S41). Significant interactions were also observed for MD in the whole WM skeleton, SCR, FXST, SS, CGC, CGH, CR, EC, PCR and SLF. In addition, significant interactions were observed in the SCR and the SLF for RD. These diagnosis-by-sex interactions were not present when we investigated adult participants (Table S42) and adolescent participants (Table S43) separately.

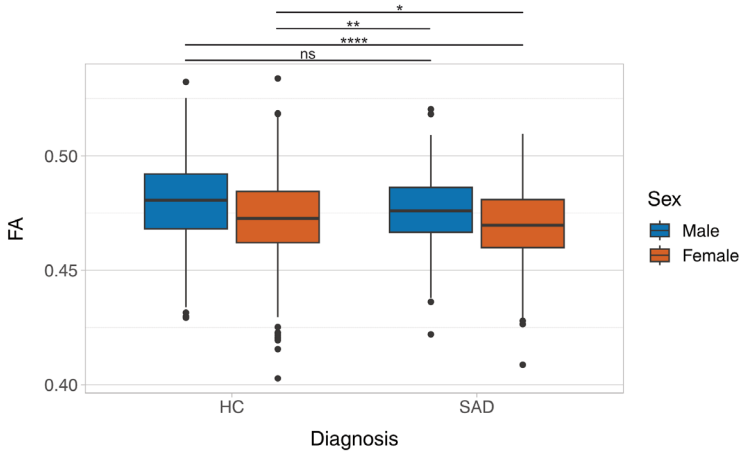



**Figure 3 Sex-by-diagnosis interactions in average fractional anisotropy (FA) over all tracts included in the present study between individuals with social anxiety disorder (SAD) and healthy controls (HC) in the full sample.** *Notes:* HC male: n = 651; HC female: n = 967; SAD male: n = 178; SAD female: n = 309. Significance after false-discovery rate adjustment for multiple comparisons: ns = not significant; * $p \leq 0.05$; ** $p \leq 10^{-2}$; *** $p \leq 10^{-3}$; **** $p \leq 10^{-4}$.

**Overlap of SAD-related WM alterations with WM patterns in with other psychiatric disorders.**

When we compared the effect sizes of WM alterations in participants with SAD with those obtained in ENIGMA-studies of other psychiatric disorders, being OCD, PTSD, schizophrenia, MDD and bipolar disorder, the effect sizes in participants with SAD were lower than these reported in these conditions (all beta < 1). There were, however, remarkable similarities between the patterns of WM alterations (Figure 4). The highest correlation was observed between the WM patterns in participants with SAD and participants with schizophrenia ($r = 0.68$, $p < 0.001$; data from [28]). In addition, significant correlations were observed for participants with MDD ($r = 0.52$, $p = 0.009$; data from [22]) and bipolar disorder ($r = 0.49$, $p = 0.015$; data from [29]). There were no significant correlations between SAD and PTSD or SAD and OCD. Detailed results are reported in Table S44.

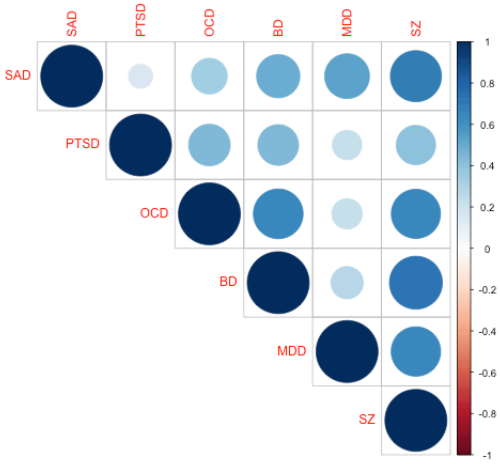



**Figure 4 Graphical depiction of correlation analyses between effect sizes of white matter microstructure in social anxiety disorder and five psychiatric disorders.** *Abbreviations:* SAD: social anxiety disorder; PTSD: post-traumatic stress disorder; OCD: obsessive compulsive disorder; MDD: major depressive disorder; SZ: schizophrenia disorder. Color bar represents Pearson *r* correlation coefficient.

## Discussion

This present study by the ENIGMA-Anxiety Working Group used a mega-analytic approach to investigate WM microstructure in adults and adolescents with SAD compared to healthy participants, combining data from twelve cohorts from research sites worldwide. We found several differences in WM microstructure between individuals with SAD and healthy controls, in the largest sample available for analysis to date. More specifically, individuals with SAD had lower average FA over all WM regions and lower FA in several regions of interest (ROIs): the corpus callosum, fornix, corona radiata and superior fronto-occipital fasciculus. A similar pattern was found when analyses were restricted to adult participants (age > 21 years). In addition, we found multiple significant interactions between sex and diagnosis as well as between age and diagnosis across the full sample. In most regions, these interactions with sex seem to be driven by lower FA in females with SAD, whereas interactions with age seem to be driven by differences in FA in adolescents with SAD. Thus, our results indicate WM differences between individuals with SAD and healthy controls as well as moderation by sex- and age-specific changes in FA in individuals with SAD.

When investigating additional parameters of WM microstructure in significant ROIs, lower FA was coupled with higher RD and MD in analyses restricted to the adult sample and in some sensitivity analyses in the full sample. Current literature suggests that this coupling could indicate demyelination [34]. However, the precise role and interpretation of additional WM parameters in SAD remains to be further investigated, as previous studies on WM in SAD report contradictory results on these parameters [7, 9, 10, 12].

As the largest WM region of the brain, the corpus callosum facilitates interhemispheric communication by connecting cortical and subcortical areas, involved in memory, attention, language, emotional states and intelligence, across the hemispheres [35]. Located at the center of the limbic system, the fornix structurally connects brain regions such as thalamus, nucleus accumbens and hippocampus that are important for emotion regulation, memory performance and reward processing [36, 37]. Notably, lower FA in the corpus callosum and fornix has been previously reported in studies on other psychiatric disorders, among others in work from ENIGMA groups on adults with MDD, bipolar disorder and schizophrenia [22, 28, 29]. However, our findings in the corpus callosum and fornix differ from findings in previous studies on WM in SAD, which did not show lower FA in these regions but reported lower FA in regions such as the UF [7-9].

As part of the ENIGMA consortium, in which preprocessing and analytical pipelines are harmonized [38], this mega-analysis allowed us to perform post-hoc investigation into the patterns of regional deficits in WM that we found in individuals with SAD relate to recent work of other ENIGMA working groups. (Figure 4). The patterns of the effect sizes of the patient-control differences in SAD showed strong correlations in regional effects sizes with three psychiatric disorders, namely schizophrenia, MDD and bipolar disorder. This overlap was observed in patterns derived from non-overlapping samples,

thus supporting the transdiagnostic nature of the observed WM deficits and underscoring the need for further exploration [39].

Our study is among the first to report widespread sex-by-diagnosis interactions in SAD, with lower FA primarily observed in females with SAD. Prior research on sex differences in WM microstructure in anxiety disorders is limited. However, a mega-analysis on preadolescent children with high and low trait anxiety reported sex-specific alterations in the uncinate fasciculus and inferior fronto-occipital fasciculus, with lower FA observed in anxious boys [40]. One study by Kim et al. [41] has reported an inverse relationship between FA and trait anxiety in right hemisphere amygdala-frontal pathways in females, while Montag and colleagues [42] found positive associations between FA and trait anxiety in left hemisphere pathways in males. This contrasts with our findings, suggesting that sex differences in WM microstructure in SAD may emerge differently in adulthood with lower FA in females with SAD. Sex differences in FA and brain ageing have been reported in the healthy population, with males typically exhibiting higher FA than females [43, 44]. However, results on sex differences in WM maturation are not uniform, and longitudinal studies in childhood, adolescence and adulthood show different trajectories for females and males [43]. These discrepancies highlight the complexity of sex-related differences in WM microstructure. Yet, as reviewed by Parsaei et al, previous studies on SAD investigated a limited number of regions in cohorts with limited age ranges [6]. This study is among the first to investigate a large number of WM regions in a substantial sample of individuals with SAD with a wide age range, thus emphasizing the need for further longitudinal investigations into how sex-specific developmental trajectories contribute to anxiety-related brain changes.

**Strengths and limitations**

Strengths of this multi-site study are the use of harmonized protocols, analyses of IPD (rather than performing meta-analyses on statistics derived from previous publications) [32] and the largest sample size of individuals with SAD to date. Furthermore, we re-used data that was previously acquired and as such DTI acquisition was not harmonized across sites. On the one hand this may have affected our findings, as DTI measurements and derived parameters are susceptible to variations in hardware [45]; on the other hand, this method variance strengthens the generalizability of our findings. Lastly, being part of the ENIGMA consortium allowed us to conduct transdiagnostic correlation analyses, which revealed overlapping WM deficits across disorders.

We also acknowledge some limitations of the present work. First, the limitations of source datasets are also applied to the pooled dataset, including cross-sectional designs and missing information about age-of-onset or ethnicity for most samples, thus disallowing causal inference and additional sensitivity analyses. Second, we used a cutoff of 21 years to categorize adolescent and adult samples. Although this is in line with previous ENIGMA studies [20, 22], alternative definitions of adolescence and adulthood could affect our findings. Third, although we corrected our analyses for site effects, a selection bias might be present as most adult studies were conducted in Europe, specifically Germany, whereas most of the adolescent samples were of North American origin. Thus, findings in the adult samples only might not be

generalizable to populations in other continents. Fifth, by averaging each ROI over the two hemispheres, we were note in the position to evaluate the precise location of findings and laterality effects. Future studies are needed to investigate voxelwise alterations in significant regions, explore generalizability of our results to other internalizing disorders and examine the role of sex on WM alterations in SAD.

## Conclusion

In this first large-scale mega-analysis investigating WM microstructure in SAD, several differences in WM regions were found between individuals with SAD and healthy controls in the full sample and in adults and adolescents separately. Our findings suggest that some WM abnormalities in SAD may not be disorder-specific but relate to transdiagnostic mechanisms underlying psychopathology. The observed sex-by-diagnosis interactions underscore the need for future research to investigate sex-specific neurodevelopmental and neurobiological factors in anxiety disorders.

**Supplemental Note S1 Deviations and Additions to the Preregistered Analyses.**

The following analyses reflect minor deviations from the preregistered plan (https://osf.io/5ycag/), made to better align with the observed data structure. First, harmonization of data was not originally included in the preregistration, as the package was still under development and ready for use a few months after preregistration. Second, correction for multiple comparisons was too strict when using the pre-registered Bonferroni method. After careful consideration, we set the threshold for significance at a false-discovery rate (FDR) corrected $p < 0{\cdot}05$, adjusting for 25 WM regions in all analyses, in line with previous ENIGMA-DTI projects [22]. Third, we only present results from additional white matter parameters, being axial, radial and mean diffusivity, if significant changes FA was found in the respective region, to avoid inflation of Type I errors and because the interpretation of these parameters is often dependent on the presence of corresponding FA alterations. Fourth, we explored sex-specific and age-specific changes only after significant effects were found in sex-by diagnosis and age-by-diagnosis analyses in several regions. Fifth, we performed correlation analyses between mega-analytical effect sizes observed in this study and the regional patterns of patient-control deficits derived using the ENIGMA-DTI workflow for five other neuropsychiatric illnesses distributed as part of the regional vulnerability index (RVI) package (https://cran.r-project.org/web/packages/RVIpkg/index.html).

**Note S2 Systematic review of mega-analytic results for comparisons in SAD vs HC and relations with comorbidity in SAD subgroups.**

***Mega-analytic results for comparisons in SAD vs HC***

After FDR correction, significant lower FA in average FA over all tracts, the BCC, CC, CR, FX, and SFO was observed in SAD patients ($n$ = 487) compared to HC ($n$ = 1604) in the full sample (Table S5). In these tracts, AD, MD or RD were further explored, but these additional WM parameters did not show significant group differences. These FA findings remained in three sensitivity analyses, investigating subsets of patients with a current SAD diagnosis ($n$ = 250), without comorbid OCD ($n$ = 447) or without comorbid PTSD ($n$ = 424; Tables S6-8).

Significantly lower FA was observed in adult patients with SAD ($n$ = 230), compared to adult HC ($n$ = 1301) in the whole WM skeleton, BCC, CC, CGC, CR, FX, GCC, PTR, SFO (Table S9). Follow-up analyses revealed higher RD in the BCC and CC. No significant effects were found for MD or AD. No significant differences were observed for FA in the full analysis between adolescent patients with SAD ($n$ = 255) patients and HC ($n$ = 292) in the same age-range (8 - 21 years old, Table S10).

***Social anxiety disorder subgroups: relations with comorbidity***

In the full sample, SAD patients with comorbid anxiety disorders with ($n$ = 142) showed lower FA in the SFO compared to healthy controls ($n$ = 1490; Table S11). Furthermore, SAD patients without comorbid anxiety disorders ($n$ = 317) displayed lower FA in the BCC and CC compared to healthy controls ($n$ = 1559; Table S12). Follow-up analyses on AD, RD or MD for in these tracts did not show any significant effects. We found no significant effects when considering SAD patients with comorbid MDD ($n$ = 184) or without comorbid MDD ($n$ = 277; Tables S13-14).

Within the adult sample, several subgroups revealed lower FA in SAD patients when compared to HCs. Specifically, patients without comorbid anxiety disorders ($n$ = 170) showed lower FA compared to HCs ($n$ = 1277) in average FA, the BCC, CC, and GCC, coupled with higher RD in these regions and higher MD in the BCC only (Table S16). Additionally, SAD patients with comorbid MDD ($n$ = 130) showed lower FA in the BCC, FX and PTR compared to HC ($n$ = 1245; Table S17). No significant effects were observed for RD, AD or MD. There were no significant differences in FA between SAD patients with comorbid anxiety disorders ($n$ = 36) or without comorbid MDD ($n$ = 98) and HCs (Tables S15,18).

No significant effects were found when considering the adolescent participants (Tables S19 – 22).

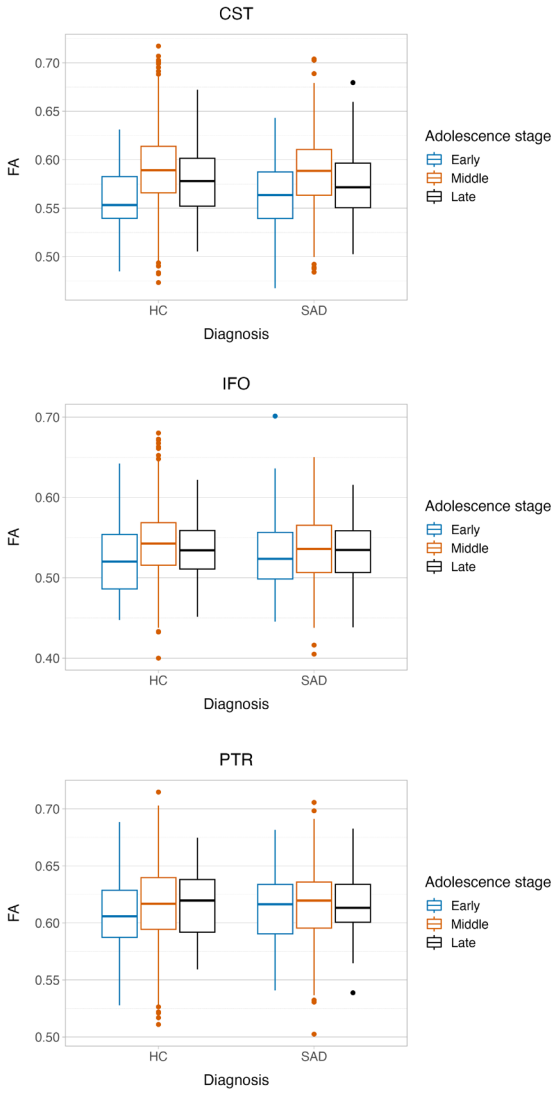
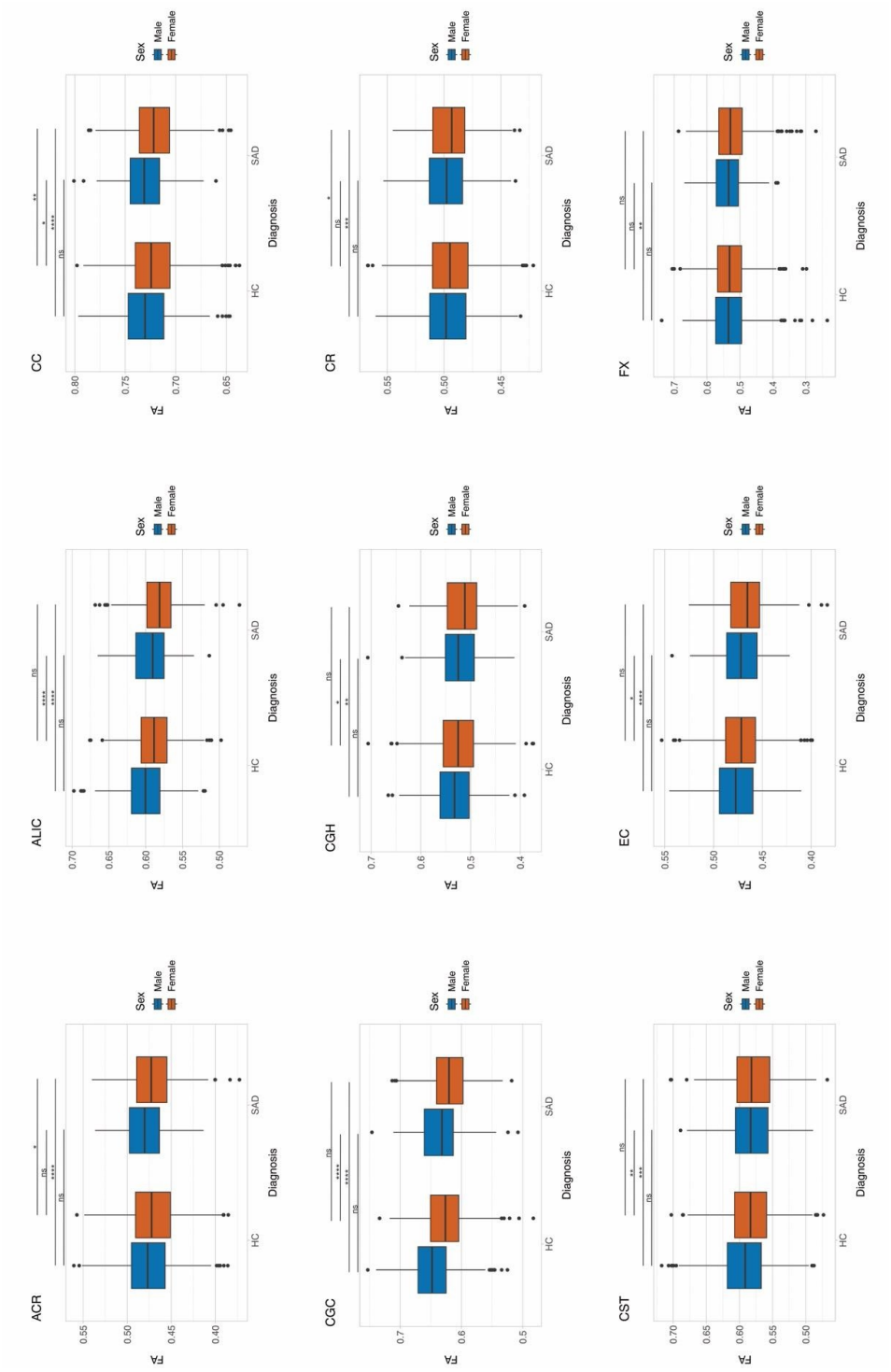



**Figure S1 Post-hoc plots illustrating diagnosis-by-age interactions when comparing adolescent patients with SAD (n = 257) to HC participants (n = 317).** *Abbreviations:* FA: fractional anisotropy; HC: healthy control; SAD: social anxiety disorder. *Note:* Adolescence stage based on age. Early: ≤ 14 years old. Middle: > 14 and < 18 years old. Late: ≥ 18 and ≤ 21 years old.


ACR
ALIC
CC
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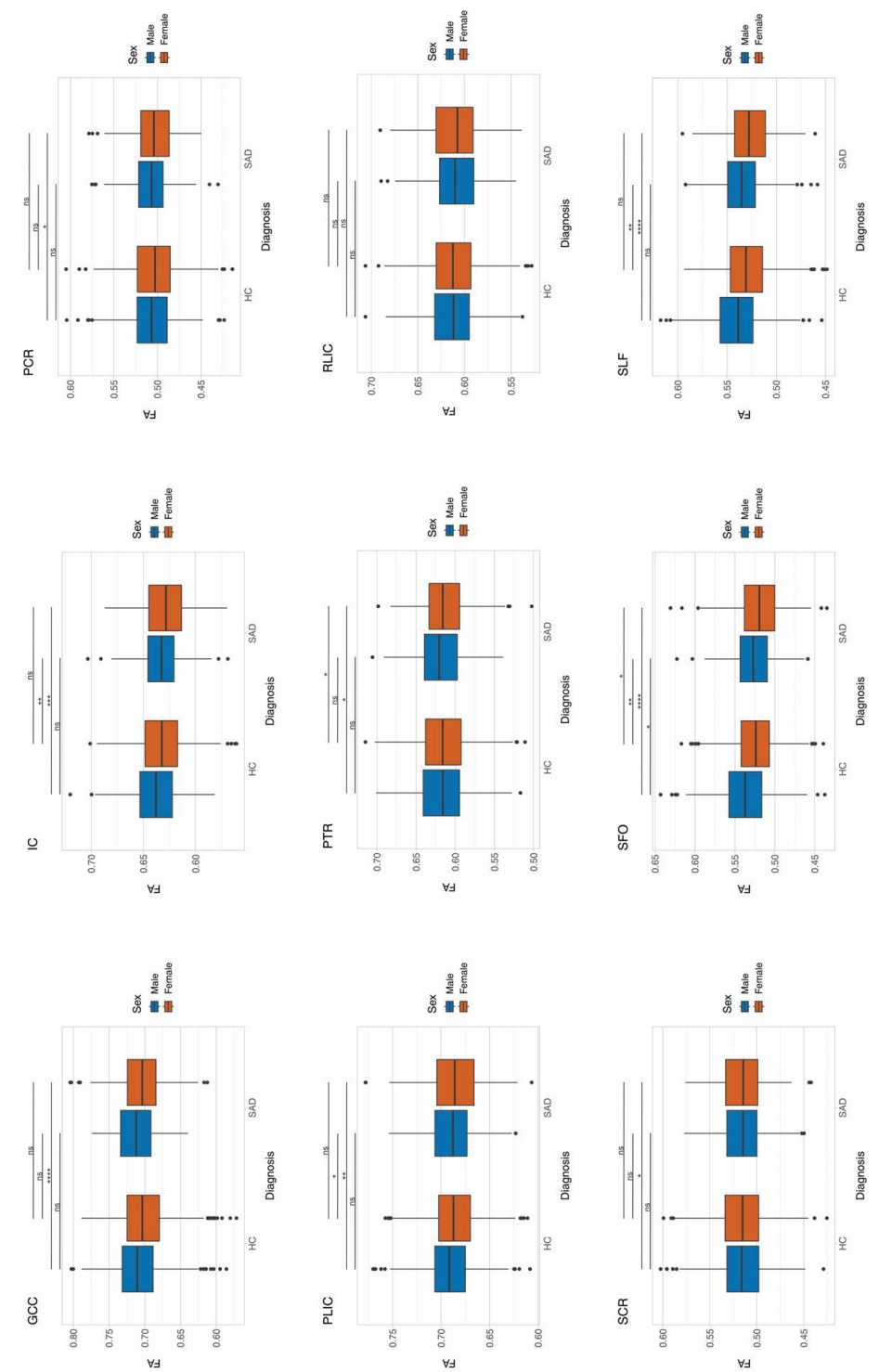


**Figure S2 Post-hoc plots illustrating significant diagnosis-by-sex interactions when comparing patients with SAD (n = 487) to HC participants (n = 1604) in the full sample.** *Note:* HC male: n = 651; HC female: n = 967; SAD male: n = 178; SAD female: n = 309. Significance after false-discovery rate (FDR) adjustment for multiple comparisons: ns = not significant; * $p \leq .05$; ** $p \leq 10^{-2}$; *** $p \leq 10^{-3}$; **** $p \leq 10^{-4}$.

**Table S1 Study information per sample on assessment measures used for SAD diagnosis and exclusion criteria.**

| Site | | Instrument | Exclusion criteria |
|---|---|---|---|
| 1 | Istanbul | SCID-CV | 1) any current psychiatric disorder other than SAD diagnosed with the SCID-I/CV;<br>2) history of alcohol or drug abuse/dependence;<br>3) any serious concomitant general medical condition or neurologic disease;<br>4) pregnancy or lactation.<br><br>Exclusion criteria for the patients and control subjects also included any contraindication for magnetic resonance imaging, and any history of neurodegenerative disease, seizure, central nervous system infection, cerebrovascular disease, diabetes mellitus, and head trauma causing loss of consciousness that lasted more than 30 min or that required hospitalization. |
| 2 | LFLSAD | MINI or MINI-Kid | Both groups: general MRI contra-indications.<br>Controls: any psychiatric diagnosis.<br>Patients: any other psychiatric diagnosis other than internalizing disorders. |
| 3 | FOR2107-MR | SCID | Any MRI contraindications; any neurological abnormalities.<br>Controls: any current or former psychiatric disorder.<br>Patients: substance dependence or current benzodiazepine treatment (wash out of at least three half-lives before study participation). |
| 4 | FOR2107-MS | SCID | Any MRI contraindications; any neurological abnormalities.<br>Controls: any current or former psychiatric disorder.<br>Patients: substance dependence or current benzodiazepine treatment (wash out of at least three half-lives before study participation). |
| 5 | Muenster | SCID | Psychotropic medication, presence or history of neurological, psychotic or bipolar disorders, drug dependence or abuse within the last 10 years, suicidal ideations, and fMRI contraindications. |
| 6 | MNC | SCID | Both groups: any neurological abnormalities, MRI contra-indications, presence or history of major internal or neurological disorder, dependence on or recent abuseof alcohol or drugs, hypertension, and general MRI contraindications.<br><br>Patients: presence of bipolar disorder, schizoaffective disorders and schizophrenia; substance related disorders or current benzodiazepine treatment (wash out of at least three half-lives before study participation), and former electroconvulsive therapy.<br><br>Controls: any current or former psychiatric disorder. |
| 7 | TIP | SCID | Both groups: any MRI contraindications; any neurological abnormalities.<br>Controls: any current or former psychiatric disorder.<br>Patients: substance dependence or current benzodiazepine treatment (wash out of at least three half-lives before study participation. |
| 8a | NIMH-SDAN | KSADS/SCID | - |

**Table S1** (continued)

| | | | |
|---|---|---|---|
| 9 | PNC | GOASSESS | (a) unable to provide signed informed consent (for participants under age 18 assent and parental consent were required);<br>(b) no English proficiency;<br>(c) physically and cognitively unable to participate in an interview and computerized neurocognitive testing. |
| 10 | MRC-SU | SCID | Any psychiatric comorbidity. |
| 11 | Vanderbilt | SCID | Both groups: unable to pass MRI safety screen, psychoactive medications in past 6 months, brain trauma.<br>Controls: any psychiatric diagnosis.<br>Patients: any other psychiatric diagnosis other than anxiety disorders. |
| 12 | West China Hospital | SCID | (1) the existence of a neurological disorder or other axis I psychiatric disorders;<br>(2) axis II antisocial or borderline personality disorders (identified using the Structured Clinical Interview for DSM-IV criteria);<br>(3) a history of drug dependence or abuse;<br>(4) pregnancy;<br>(5) major physical illness such as cardiovascular disease or hepatitis, as assessed by clinical evaluations and medical records;<br>(6) any other DSM-IV axis I comorbidity. |

*Abbreviations:* SCID-CV: Structured Clinical Interview for DSM-5 Disorders–Clinician Version. MINI: Mini-International Neuropsychiatric Interview. MINI-Kid: Mini-International Neuropsychiatric Interview for Children and Adolescents (MINI-KID). KSADS: Schedule for Affective Disorders and Schizophrenia for School-Aged Children (K-SADS). GOASSESS: Grand Opportunity Assessment [46]. SCID: Structured Clinical Interview for DSM-5 Disorders. *Notes:* Information not received (-).

**Table S2 Detailed clinical characteristics of the cohorts included for mega-analysis (selectively reported for social anxiety disorder patients).**

| | Site | Comorbid psychiatric disorders (%) | | | | | | | | | Medication use (%) | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | PD | AG | GAD | SPH | Other-ANX | MDD | OCD | PTSD | OtherDx | Benzo | Oth-erMed |
| 1 | Istanbul | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| 2 | LFLSAD | 30.0 | 10.0 | 10.0 | 10.0 | 0.0 | 50.0 | 10.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| 3 | FOR2107-MR | 7.0 | 7.0 | 7.0 | 15.8 | 0.0 | 98.2 | 0.0 | 19.3 | 8.8 | 0.0 | 24.6 |
| 4 | FOR2107-MS | 19.4 | 19.4 | 12.9 | 3.2 | 0.0 | 96.8 | 6.5 | 3.2 | 29.0 | 0.0 | 32.3 |
| 5 | Muenster | NA | NA | NA | NA | NA | NA | NA | NA | NA | 0.0 | 0.0 |
| 6 | MNC | 3.4 | 0.0 | 0.0 | 8.6 | 0.0 | 46.6 | 1.7 | 3.4 | 1.7 | 0.0 | 1.7 |
| 7 | TIP | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 82.1 | 0.0 | 0.0 | 0.0 | 0.0 | 5.1 |
| 8 | NIMH-SDAN | 0.0 | 20.8 | 66.7 | 0.0 | 41.7 | 0.0 | 0.0 | 0.0 | 18.2 | 0.0 | 0.0 |
| 9 | PNC | 0.5 | 7.6 | 3.3 | 34.2 | 7.0 | 15.1 | 4.3 | 12.1 | 41.6 | 0.0 | 0.0 |
| 10 | MRC-SU | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 50.0 | 0.0 | 0.0 | NA | 0.0 | 0.0 |
| 11 | Vanderbilt | 0.0 | 33.3 | 16.7 | 50.0 | 0.0 | 0.0 | 33.3 | 16.7 | 0.0 | 0.0 | 0.0 |
| 12 | West China Hospital | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |

*Abbreviations:* PD: Panic Disorder; AG: Agoraphobia; GAD: Generalized Anxiety Disorder; SPH: Specific Phobias; OtherANX: Other comorbid anxiety disorder; OCD: Obsessive-Compulsive Disorder; PTSD: Post-Traumatic Stress Disorder; OtherDx: Other comorbid psychiatric disorder (does not include major depressive disorder); Benzo: Benzodiazepines; OtherMed: Other psychotropic medication (does not include selective serotonin reuptake inhibitors or serotonin-norepinephrine reuptake inhibitors); NA: not available (information not recorded in this sample).

**Table S3 Image acquisition in the cohorts included for mega-analysis.**

| Site | | Scanner type | Field strength | Voxel size | Slice thickness | Number of directions & b-factor |
|---|---|---|---|---|---|---|
| 1 | Istanbul | Philips Achieva | 1.5T | 1.75x1.75x2mm | 2mm | 32 directions, b-factor = 800, 1 b0 image |
| 2 | LFLSAD | Philips Achieva | 3T | 1.9 x 2.4mm | 2mm | 30 directions, b-factor = 1000, 2 b0 images |
| 3 | FOR2107-MR | Siemens Magnetom Trio | 3T | 2.5 x 2.5 mm | 2.5 mm | 30 directions; 4b images; b-factor = 1000 |
| 4 | FOR2107-MS | Siemens Prisma | 3T | - | - | - |
| 5 | Muenster | Siemens Magnetom PRISMA | 3T | 2.3x2..3x3 mm | 3mm | 30 directions, b-factor = 1000, 4 b0 images |
| 6 | MNC | Philips Gyroscan Intera | 3T | - | - | - |
| 7 | TIP | - | - | - | - | - |
| 8a | NIMH-SDAN | GE MR 750 | 3T | 1.875mm$^3$ | 2.5mm | 62 directions, b-factor = 1000, 2 b0 images |
| 8b | PNC | GE MR 750 | 3T | 1.87mm$^3$ | 2.5mm | 75 directions, b-factor = 300 (6) and 1100 (69), 5 b0 images |
| 8c | MRC-SU | GE MR 750 | 3T | 2x2mm | 2.9mm | 48 directions, b-factor = 1000, 8 b0 images |
| 9 | Vanderbilt | Siemens Trio | 3T | 1.8x1.8x2 mm | 2mm | 64 directions, b-factor = 1000, 7 b0 images |
| 10 | West China Hospital | Siemens Magnetom Allegra | 3T | 1.8 x 1.8 x 2.0mm | 2mm | - directions, b-factor = 1000 |
| 11 | Istanbul | Philips Achieva | 3T | 2.5mm$^3$ | 2.5mm | 92 directions, b-factor = 1600, 1 b0-mage |
| 12 | LFLSAD | GE SIGNA EXCITE | 3T | 0.9375mm$^3$ | 3mm | 16 directions |

*Abbreviations:* GE: General Electric. *Notes:* Information not provided (-).

**Tables S4 – S44 Mega-analytic results and correlation analyses.**

Due to their size, these tables are available using the QR code below.

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